ÖGH Herpetozoa
Austrian Herpetological Society

Herpetozoa 39: 49–79 (2026)
DOI 10.3897/herpetozoa.39.e184954

A new treefrog of the genus *Charadrahyla* (Anura, Hylidae) from Oaxaca, Mexico, with comments on the conservation status and distribution of related species

Luis Canseco-Márquez[1], Christoph I. Grünwald[2,3,4], Jacobo Reyes-Velasco[3,4], Carlos Montaño-Ruvalcaba[3,4], Jonathan A. Campbell[5]

1 *Museo de Zoología, Departamento de Biología Evolutiva, Facultad de Ciencias, Universidad Nacional Autónoma de México, Cd. Universitaria, Del. Coyoacán, Mexico 04510, Ciudad de México, Mexico*
2 *Biencom Real Estate, Carretera Chapala–Jocotepec #57-1, C.P. 45920, Ajijic, Jalisco, Mexico*
3 *Biodiversa A.C., Avenida de la Ribera #203, C.P. 45900, Chapala, Jalisco, Mexico*
4 *Herp.mx A.C. C.P. 28989, Villa de Álvarez, Colima, Mexico*
5 *Department of Biology, University of Texas at Arlington, Arlington, Texas 76010, USA*

https://zoobank.org/4BF1A241-72FA-4268-869D-DD8C7490DD88

Corresponding author: Christoph I. Grünwald (cgruenwald@switaki.com)

Academic editor: Arthur Tiutenko ♦ **Received** 12 January 2026 ♦ **Accepted** 13 March 2026 ♦ **Published** 27 March 2026

Abstract

We describe a new species of *Charadrahyla* from the Sierra Juárez portion of the Sierra Madre Oriental in the state of Oaxaca, Mexico, and provide morphological and molecular data to support the novelty of this species. The new species is most similar to *Charadrahyla nephila*, with which it has been confused in the past. We report on adult, larval, and juvenile specimens of this new species and parapatric *C. nephila*. We also report a novel population of the endangered *Charadrahyla sakbah* from the Sierra Monteflor portion of the Sierra Madre Oriental, also in Oaxaca, which significantly extends its known distribution. We provide an updated distribution map for species of *Charadrahyla* inhabiting Oaxaca. Finally, we provide conservation recommendations for various species in this genus, at least four of which should be considered Endangered.

Resumen

Describimos una nueva especie de *Charadrahyla* de la Sierra Juárez, en la Sierra Madre Oriental, Oaxaca, México, y proporcionamos datos morfológicos y moleculares que respaldan la distinción de esta especie. Esta nueva especie está cercanamente relacionada con *Charadrahyla nephila*, con la cual se ha confundido en el pasado. Informamos sobre especímenes adultos, larvas y juveniles de esta especie y de *C. nephila*. También presentamos una nueva población de *Charadrahyla sakbah*, una especie considerada en peligro de extinción, de la Sierra Monteflor, también en la Sierra Madre Oriental de Oaxaca, la cual extiende su distribución significativamente. Además, proporcionamos un mapa de distribución actualizado para las especies de *Charadrahyla* en Oaxaca. Finalmente, ofrecemos recomendaciones de conservación para las diversas especies de *Charadrahyla*, de las cuales consideramos que al menos cuatro deberían considerarse en peligro de extinción.

Copyright *Luis Canseco-Márquez et al.* This is an open access article distributed under the terms of the Creative Commons Attribution License (CC BY 4.0), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

Kurzfassung

Wir beschreiben eine neue Nebelwald-bewohnende Art von *Charadrahyla* aus der Sierra Juárez in der Sierra Madre Oriental im Bundesstaat Oaxaca, Mexiko, und legen morphologische und molekulare Daten vor, die die Hypothese dass es sich um eine neue Art handelt unterstützen. Die neue Art ist am nächsten mit *Charadrahyla nephila* verwandt, mit der sie in der Vergangenheit öfters verwechselt wurde. Wir berichten über Erwachsene, Larven und Jungtiere dieser Art und der nahverwandten *C. nephila*. Außerdem berichten wir über einen neuen Fundort des gefährdeten *Charadrahyla sakbah* aus der Sierra Monteflor in der Sierra Madre Oriental, ebenfalls in Oaxaca und wir erweitern sein Verbreitungsgebiet erheblich. Wir fügen eine aktualisierte Verbreitungskarte von *Charadrahyla* in Oaxaca bei. Abschließend geben wir Empfehlungen zum Artenschutz für die verschiedenen Arten von *Charadrahyla*, von denen unserer Ansicht nach mindestens vier als gefährdet gelten sollten.

Key Words

Amphibia, conservation, IUCN, phylogeny, range map, Sierra Madre Oriental, systematics, taxonomy

Palabras clave

Amphibia, conservación, filogenia, IUCN, mapas de distribución, Sierra Madre Oriental, sistemática, taxonomía

Schlüsselwörter

Amphibien, Artenschutz, IUCN, Phylogenie, Sierra Madre Oriental, Systematik, Taxonomie, Verbreitungskarte

Introduction

Treefrogs of the genus *Charadrahyla* Faivovich, Haddad, Garcia, Frost, Campbell & Wheeler, 2005 are endemic to Mexico and inhabit tropical and temperate forests on the Pacific Versant of Guerrero and Oaxaca and the Atlantic Versant on the eastern flanks of the Sierra Madre Oriental from San Luis Potosí (Escobedo-Cadena et al. 2024) to Chiapas. Currently, ten species are recognized in the genus *Charadrahyla*, several of which are considered to be at imminent risk of extinction (Endangered, IUCN 2025). While the different species remain extant, individuals from various populations of these frogs have not been observed or collected in half a century or more and may be locally extinct (see below). Duellman first identified a *Hyla taeniopus* Group (Duellman, 1970) within which he placed *Charadrahyla altipotens* (Duellman, 1968), *Charadrahyla taeniopus* (Günther, 1901), *Charadrahyla chaneque* (Duellman, 1961) and *Charadrahyla duellmani* (Lynch & Smith, 1966), albeit the latter as a synonym of *C. chaneque*. Two more species, *Charadrahyla trux* (Adler & Dennis, 1972) and *Charadrahyla nephila* (Mendelson & Campbell, 1999) were described later. Faivovich et al. (2005) erected the genus *Charadrahyla* for the species formerly considered to belong to the *Hyla taeniopus* Group. It should be noted that they diagnosed the genus by molecular divergences between species, without morphological synapomorphies, and only included *C. nephila* and *C. taeniopus* in their analysis. Recently, three more species have been described in the genus *Charadrahyla*; *Charadrahyla tecuani* Campbell, Blancas-Hernández and Smith (2009), *Charadrahyla esperancensis* Canseco-Márquez et al. (2017), and *Charadrahyla sakbah* Jiménez-Arcos et al. (2019). Further, in a broad phylogenetic study of the Hylidae, Faivovich et al. (2018) transferred *Hyla pinorum* Taylor (1937) and *Hyla juanitae* (Snyder, 1972), formerly placed in *Exerodonta*, to the genus *Charadrahyla* based on molecular data, with high support.

Recent surveys in the Sierra Madre Oriental of Oaxaca have produced several specimens of an arboreal treefrog similar to *C. chaneque* but not clearly assignable to that species. The smaller adult size, extremely tuberculate dorsal skin, unique color pattern, distinct eye coloration, and other discrete morphological traits readily differentiate this novel species from its congeners. We describe this species below.

Materials and methods

Taxonomic sampling

Numerous trips were made in southern Mexico between 2008 and 2024 to collect specimens of *Charadrahyla*. We euthanized frogs with topical application of 1% benzocaine and collected tissue samples from thigh muscle

upon death and preserved them in 96% ethanol. Specimens were fixed in a 10% stock formalin solution and subsequently stored in 70% ethanol. All material collected was deposited in the herpetological collection of the Museo de Zoología, Facultad de Ciencias (MZFC) and the Colección Nacional de Anfibios y Reptiles, Instituto de Biología (CNAR), both located at the Universidad Nacional Autónoma de Mexico (UNAM) in Mexico City, Mexico; and the Instituto de Investigaciones sobre los Recursos Naturales (INIRENA) of the Universidad Michoacana de San Nicolás de Hidalgo (UMSNH) in Morelia, Michoacán, Mexico (See Appendix 1).

Material examined

We examined and measured specimens of *Charadrahyla* housed at MZFC and CNAR. We also examined photographs of specimens housed at: Amphibian and Reptile Diversity Research Center of the University of Texas at Arlington (UTA); University of Illinois, Museum of Natural History (UIMNH); University of Kansas Natural History Collection (KU); University of Colorado Museum of Natural History (UCM). Field number abbreviations are as follows: CIG (Christoph I. Grünwald); ANMO (Adrián Nieto Montes de Oca); JAC (Jonathan A. Campbell); LCM (Luis Canseco-Márquez): ANMO and LCM to be deposited in MZFC; JAC in UTA; CIG in either MZFC or INIRENA. Specimen numbers for all material examined, including specimens examined by photographs, are provided in Appendix 1. Museum acronyms throughout follow Sabaj-Pérez (2016).

We examined specimens of every known species of *Charadrahyla* (Appendix 1), or reviewed photographs of relevant specimens for comparison whenever first-hand examination was not possible (noted in Appendix 1 as "photo only").

Comparisons with *C. chaneque* are taken from Duellman (1961, 1970, 2001) and from specimens collected near the type-locality. However, Duellman (1970, 2001) based some of his description on specimens from the Chimalapas (described as *C. duellmani*). We consider these specimens of equivocal status, and a concerted effort should be made to rediscover this population to determine its relationships and conservation status. For this study, the type-specimen of *C. chaneque* was examined only through photographs.

Comparisons with *C. nephila* were made using data from Mendelson and Campbell (1999) and Duellman (2001), as well as with specimens collected by us near the type-locality and in the Sierra Juárez. We only used characters from specimens we were able to positively identify as *C. nephila* from photographs. The type-specimen of *C. nephila* was examined from photographs. We analyzed the measurements used by Mendelson and Campbell (1999) for the description of *C. nephila* (provide by J. Mendelson, pers. comm.), however, due to possible differences in methodology, we did not use that data except to help determine size ranges for adult *C. nephila* and the new species.

Distribution maps were generated based on the Global Biodiversity Information Facility database (www.gbif.org), which includes both museum records as well as distribution records from the iNaturalist platform (inaturalist.org). All identities of individual records included in the maps were verified by us using photographs. Additional localities mapped were taken from taxonomic works (Duellman 1970, 2001; Mendelson and Campbell 1999; Canseco-Márquez et al. 2017; Jiménez-Arcos et al. 2019), and specimens collected by us.

Molecular analysis

DNA extraction and PCR amplification

A detailed description of DNA extraction and PCR amplification protocols followed here is provided in Grünwald et al. (2018). We extracted genomic DNA from tissue samples using a standard potassium acetate protocol. We amplified and sequenced a fragment of the mitochondrial 16S rRNA gene using the primers LX12SN1a (forward) and LX16S1Ra (reverse) of Zhang et al. (2013), or the modified primers 16Sar and 16Sbr of Bossuyt and Milinkovitch (2000). For the mitochondrial 12S rRNA gene, we used the primers 12SAL (forward) and 16S2000H (reverse) of Zhang et al. (2013). PCR conditions followed those described by Zhang et al. (2013) Sequencing of unpurified PCR products was carried out by BGI Tech Solutions (Hong Kong).

To infer phylogenetic relationships, we included additional sequences of *Charadrahyla* and other hylid frogs obtained from GenBank and from Caviedes-Solis et al. (2020). Our dataset comprised five loci for species of *Charadrahyla* and outgroup taxa: three mitochondrial genes (16S, 12S, and ND1) and two nuclear genes (Rag-1 and rhodopsin). All sequences used in this study, along with their GenBank accession numbers, are listed in Appendix 2: Table A1.

Sequence alignment and phylogenetic analysis

We manually trimmed the 5' and 3' ends of all sequences to remove regions with poor-quality base calls using Geneious v6.1.6 (Biomatters Ltd., Auckland, New Zealand). We aligned each locus separately in MAFFT v7 (Katoh et al. 2019) using the Q-INS-I algorithm. The concatenated alignment comprised 3,699 base pairs, including 946 base pairs of 12S; 580 base pairs of 16S; 1,381 base pairs of ND1; 454 base pairs of Rag-1; and 338 base pairs of Rhodopsin. The final dataset included 18 individuals of *Charadrahyla* and five of the closely related *Megastomatohyla* Faivovich et al., 2005. We deposited all newly generated sequences in GenBank and list their accession numbers in bold in Appendix 2: Table A1.

We inferred phylogenetic relationships using maximum likelihood (ML) analyses in IQ-TREE (Nguyen et al. 2015) via its online platform (Trifinopoulos et al.

2016). We analyzed the concatenated mitochondrial and nuclear dataset as well as each locus independently. IQ-TREE selected substitution models automatically, and we estimated node support using 1,000 ultrafast bootstrap replicates. We considered clades as strongly supported if they had a UFB value of 95% or higher. We visualized the resulting topologies in FigTree v1.4.2 (Rambaut 2014).

Genetic distances

We calculated uncorrected pairwise genetic distances (p-distances) for the mitochondrial 12S and 16S rRNA genes using MEGA X (Kumar et al. 2018). We estimated distances separately for each locus based on aligned sequences and included both intra- and interspecific comparisons when multiple samples were available. We report distances as percentages.

Morphological measurements

Most characters and terminology follow Duellman (1970); additionally, we took some measurements not taken by Duellman (1970). Measurements taken for each specimen (abbreviations listed in parenthesis) are as follows: snout–vent length (SVL), head length, from the tip of the snout in a straight line to immediately above jaw angle (HL), head width at tympanum (HW), head height (HH), measured from posterior to the rear terminus of the eyelid, between the eye and tympanum; eyelid width (ELW), interorbital distance (IOD), internarial distance (IND), eye–naris distance (END), diameter of eye (ED), width of tympanum (TW), eye–tympanum distance (ETD), upper arm length, measured from axilla to elbow (UaL), forearm length, measured from elbow to proximal end of outer palmar tubercle (FaL), hand length, measured from the proximal end of the outer palmar tubercle to tip of the third finger (HaL), length of 1st finger, (1FL), length of 2nd finger (2FL), length of 3rd finger (3FL), width of pad on 3rd finger (3FPW), width of 3rd finger (3FW), length of 4th finger (4FL), width of pad on 4th finger (4FPW), width of 4th finger (4FW), femur length (FeL), tibia length (TiL), tarsal length (TaL), foot length, measured from the proximal end of the outer metatarsal tubercle to the tip of the pad on the fourth (longest) toe (FL), length of 1st toe (1TL), length of 2nd toe (2TL), length of 3rd toe (3TL), width of pad on 3rd toe (3TPW), width of 3rd toe (3TW), length of 4th toe (4TL), width of pad on 4th toe (4TPW), width of 4th toe (4TW), and length of 5th toe (5TL). Measurements taken on left hand side whenever possible, otherwise mentioned. All finger and toe lengths were measured from the proximal end of the first subarticular tubercle to the tip of the pad on the digit.

Measurements were made with Truper (Mexico) brand digital calipers and rounded to the nearest 0.1 mm. The sex of adult specimens was determined by presence of vocal slits, secondary sexual characters such as nuptial excrescences, or by having observed the males calling in life. Webbing formulae follow Myers and Duellman (1982) as modified by Savage and Heyer (1997). Descriptions of color in life are based on photographs taken of the live specimens.

Results

Molecular phylogenetic results

We obtained a total of seven novel DNA sequences of the mitochondrial 12S gene, including multiple samples from five described species of *Charadrahyla* (*C. altipotens*, *C. chaneque*, *C. juanitae*, *C. nephila*, and *C. pinorum*), as well as two samples of the new species. These sequences were analyzed together with three *Charadrahyla* and two *Megastomatohyla* 12S sequences available in Genbank (Appendix 2: Table A1). We also obtained four novel DNA sequences of the mitochondrial 16S gene, representing one individual of *C. altipotens*, one sample of the new species, and one each of *M. mixomaculata* and *M. pellita*. These sequences were analyzed along with seven *Charadrahyla* and two *Megastomatohyla* 16S sequences available in Genbank (Appendix 2: Table A1); as well as six 16S sequences published by Caviedes-Solis et al. (2020), but not available in Genbank. Finally, we included in our analysis sequences of several other genes including one mitochondrial gene (ND1) and two nuclear genes (RAG1, Rhodopsin) available in Genbank (Appendix 2: Table A1) or published by Caviedes-Solis et al. (2020).

We compared the new species with all currently recognized taxa of *Charadrahyla* for which molecular data are available, with the exception of *C. tecuani*, for which no DNA sequences exist. The absence of *C. tecuani* does not affect the present study, as the species is known only from its type locality in the Sierra Madre del Sur of Guerrero, more than 500 km west of the Sierra Madre Oriental and is diagnosably different from the species described herein.

Although the available sequence data do not allow reconstruction of a well-resolved evolutionary history for the genus *Charadrahyla*, they support the recognition of a distinct evolutionary lineage from the Sierra Juárez. Phylogenetic analyses recovered *Charadrahyla* as monophyletic and sister to the genus *Megastomatohyla*, which is endemic to the cloud forests of central Veracruz and Oaxaca. *Charadrahyla taeniopus* was recovered as sister to all remaining species in the genus with strong support (100% ultrafast bootstrap support, ufb), followed by *C. chaneque* (89% ufb). Most internal nodes within the genus received low support (<70% ufb).

The analyses recovered *C. esperancensis* as sister to *C. sakbah* (73% ufb), whereas *C. pinorum*, *C. altipotens*, and *C. juanitae* formed a well-supported clade (93% ufb). The new species was recovered as sister to *C. nephila*, although this relationship received low support (41% ufb). The included samples of our novel species were recovered in a monophyletic clade with strong support (100% ufb) (Fig. 1).

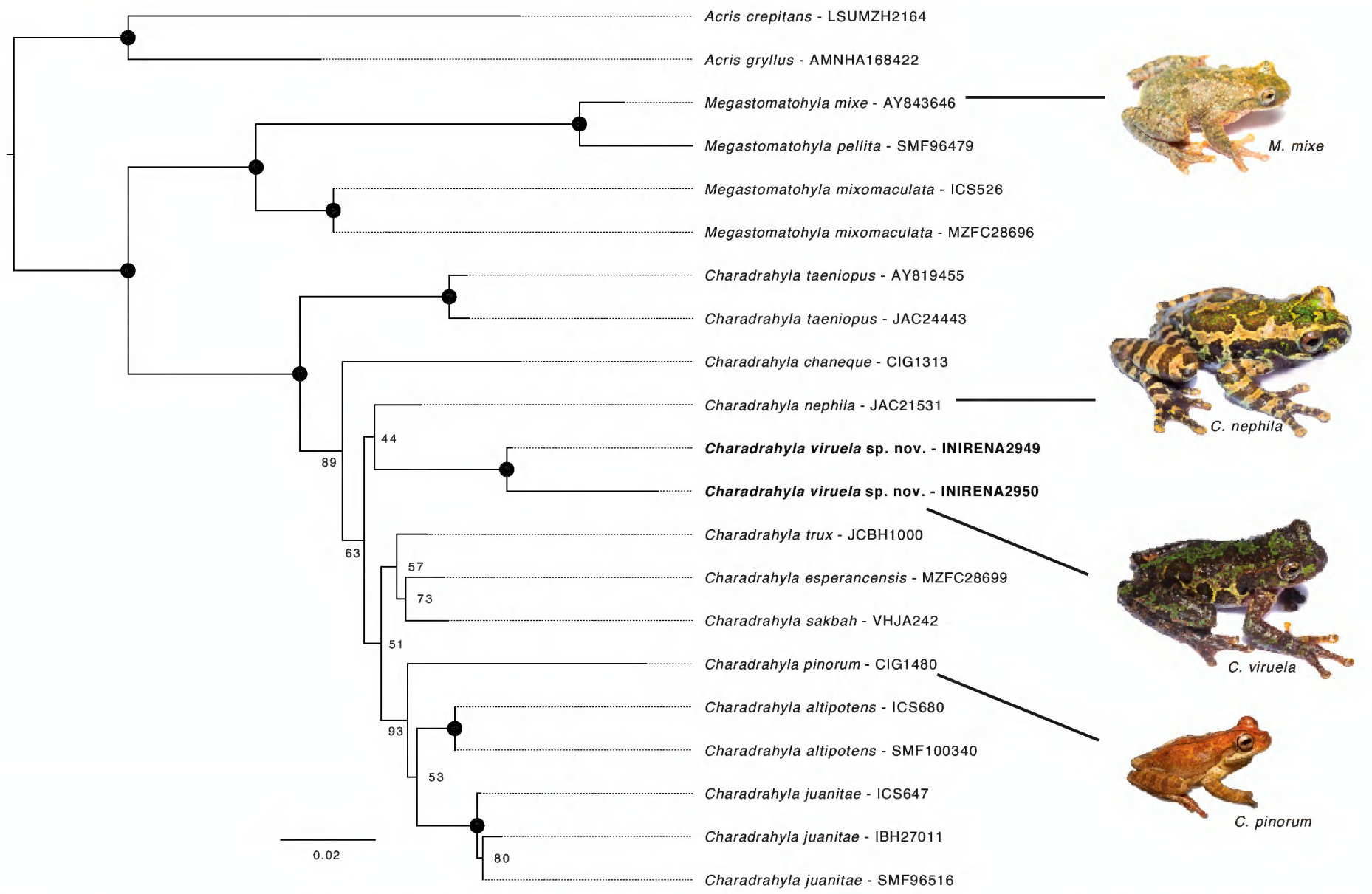

Figure 1. Maximum-likelihood phylogenetic tree of members of the frog genus *Charadrahyla* and *Megastomatohyla*, inferred from three mitochondrial loci (12s, 16s, ND1) and two nuclear loci (RAG1 and rhodopsin). Numbers at nodes indicate bootstrap support values based on 1,000 ultrafast replicates. Black circles denote nodes with 100% bootstrap support.

Genetic distances

For the 12S gene, interspecific genetic distances within *Charadrahyla* ranged from 4.1–11.8%. *Charadrahyla* sp. nov. showed its lowest divergence relative to *C. nephila* (4.1%), whereas distances to other congeners reached higher values, up to 11.8% in comparisons with *C. juanitae*. Intraspecific divergence within *C. viruela* sp. nov. were 1.6% between two samples, whereas intraspecific distances in *C. taeniopus* between three samples from two localities, both in Puebla, ranged from 0–0.7%.

For the 16S gene, interspecific genetic distances within *Charadrahyla* ranged from 2.4–6.3%. The lowest interspecific values occurred among other species pairs (e.g., *C. trux* and *C. esperancensis*, 2.4%). *Charadrahyla viruela* sp. nov. differed from *C. nephila* by 3.4%, and showed larger divergences from other congeners, reaching 6.3% in comparisons with *C. juanitae*.

Intergeneric genetic distances between *Charadrahyla* and *Megastomatohyla* ranged from 11.7–15.6% for 12S and from 9.4–11.9% for 16S. Interestingly, interspecific genetic distances within *Megastomatohyla* ranged from 1.2–3.0%, values lower than most interspecific comparisons observed within *Charadrahyla*, with 1.2% genetic distance in 16S between *M. mixe* and *M. pellita* and 2.9% genetic distance in 12S between *M. mixe* and *M. pellita*, both being the lowest interspecific genetic distances we recorded for the group.

Tadpole analysis

In our description of the tadpoles of *Charadrahyla* we refer to them in stages outlined in Gosner (1960). We refer to these stages as Gosner Stages (GS). Terminology and measurements follow Altig and McDiarmid (1999).

Systematic account

Charadrahyla viruela sp. nov.

https://zoobank.org/D4745671-9ED1-4FC9-B6B3-75BE1FBA5B87

Figs 2, 3, 4A, C, 5A, B

Hyla chaneque—Duellman (1965): 164–167, in part.
Hyla chaneque—Duellman (1970): 440–445, 699, in part.
Hyla nephila—Mendelson and Campbell (1999): 80–86, in part.
Hyla nephila—Duellman (2001): 937–939, 1128, in part.
Charadrahyla nephila—Canseco-Márquez, Ramírez-González, González-Bernal (2017): 64–72, in part.
Charadrahyla nephila—Jiménez-Arcos, Calzada-Arciniega, Alfaro-Juantorena, Vázquez-Reyes, Blair, Parra-Olea (2019): 371–385, in part.
Charadrahyla sp.—León-Gómez (2019): 1–79.

Proposed standard English name. Tuberculate Cloud-forest Treefrog

Proposed standard Spanish name. Ranita Verrugosa de Árbol de Bosque Mesófilo

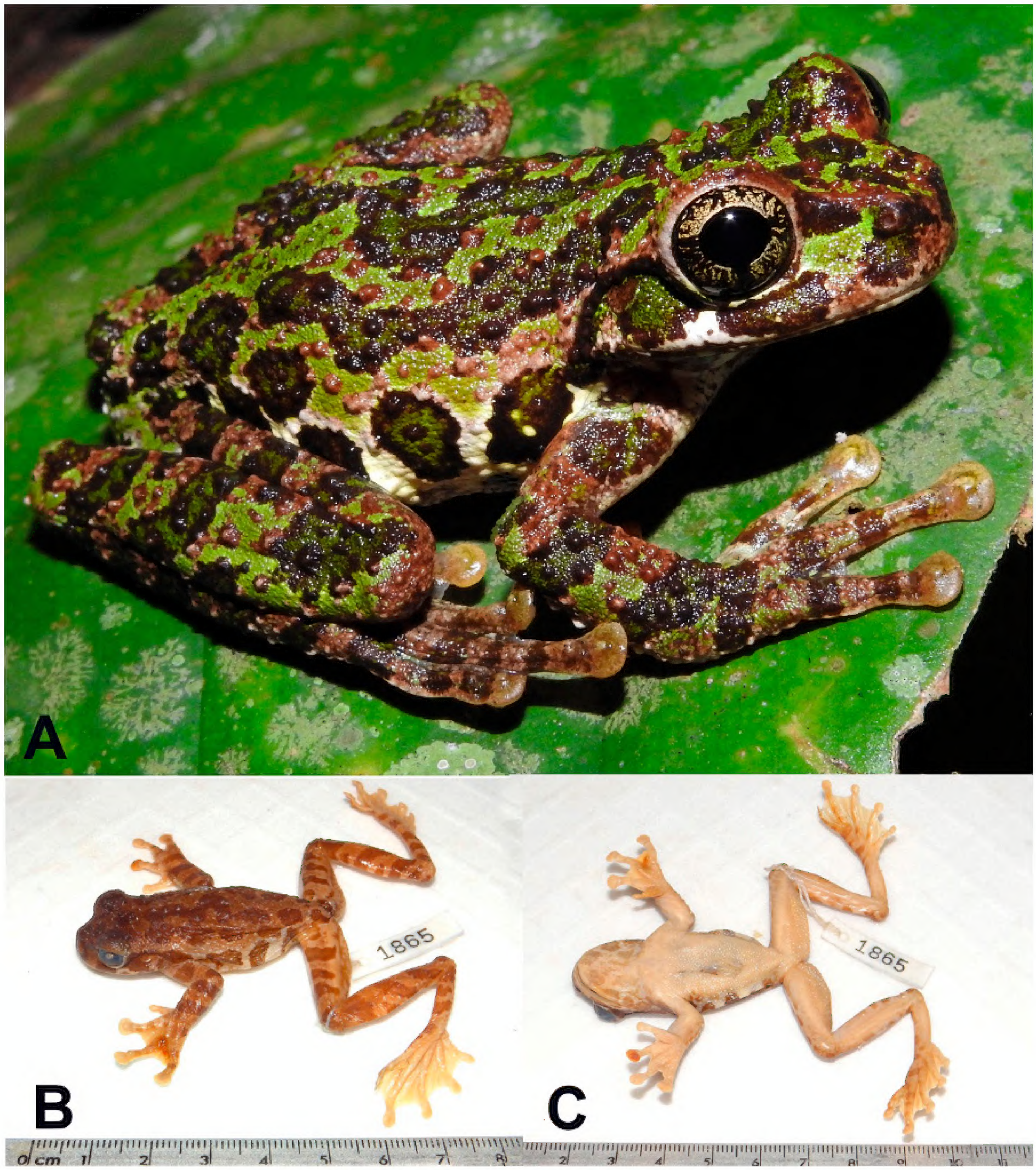

Figure 2. *Charadrahyla viruela* sp. nov. Holotype (**INIRENA 2950**). Live specimen (**A**) and preserved in dorsolateral (**B**) and ventrolateral (**C**) views.

Holotype. INIRENA 2950 (CIG 1865). Subadult female (Fig. 2) from Highway 175 between Vista Hermosa and La Esperanza, 1.9 km ENE of La Esperanza, Municipio de Santiago Comaltepec (17.6264, -96.3520; 1,589 m asl; datum = WGS84), Oaxaca, Mexico, collected on 06 May 2021 by Christoph I. Grünwald, André J. Grünwald, and Iván Ahumada-Carrillo.

Paratypes (n = 6, Fig. 3). MZFC 37345 (LCM 1700), an adult female, Arroyo Tres Manantiales, La Esperanza, Municipio de Santiago Comaltepec (17.6236, -96.3654, 1,688 m asl; datum = WGS84), Oaxaca, Mexico, collected on 13 March 2017 by Luis Canseco-Márquez and Cynthia G. Ramírez-González; **MZFC 37344** (CIG 1172), an adult male from Highway 175 at La Esperanza, Municipio de Santiago Comaltepec (17.6229, -96.3656, 1,675 m asl; datum = WGS84), Oaxaca, Mexico, collected on 25 July 2017 by Héctor Franz-Chávez, André J. Grünwald, and Christoph I. Grünwald; **MZFC 15974–75**, 2 adult males, 4.2 km S of Vista Hermosa, and approx. 1.2 km S of La Esperanza, Municipio de

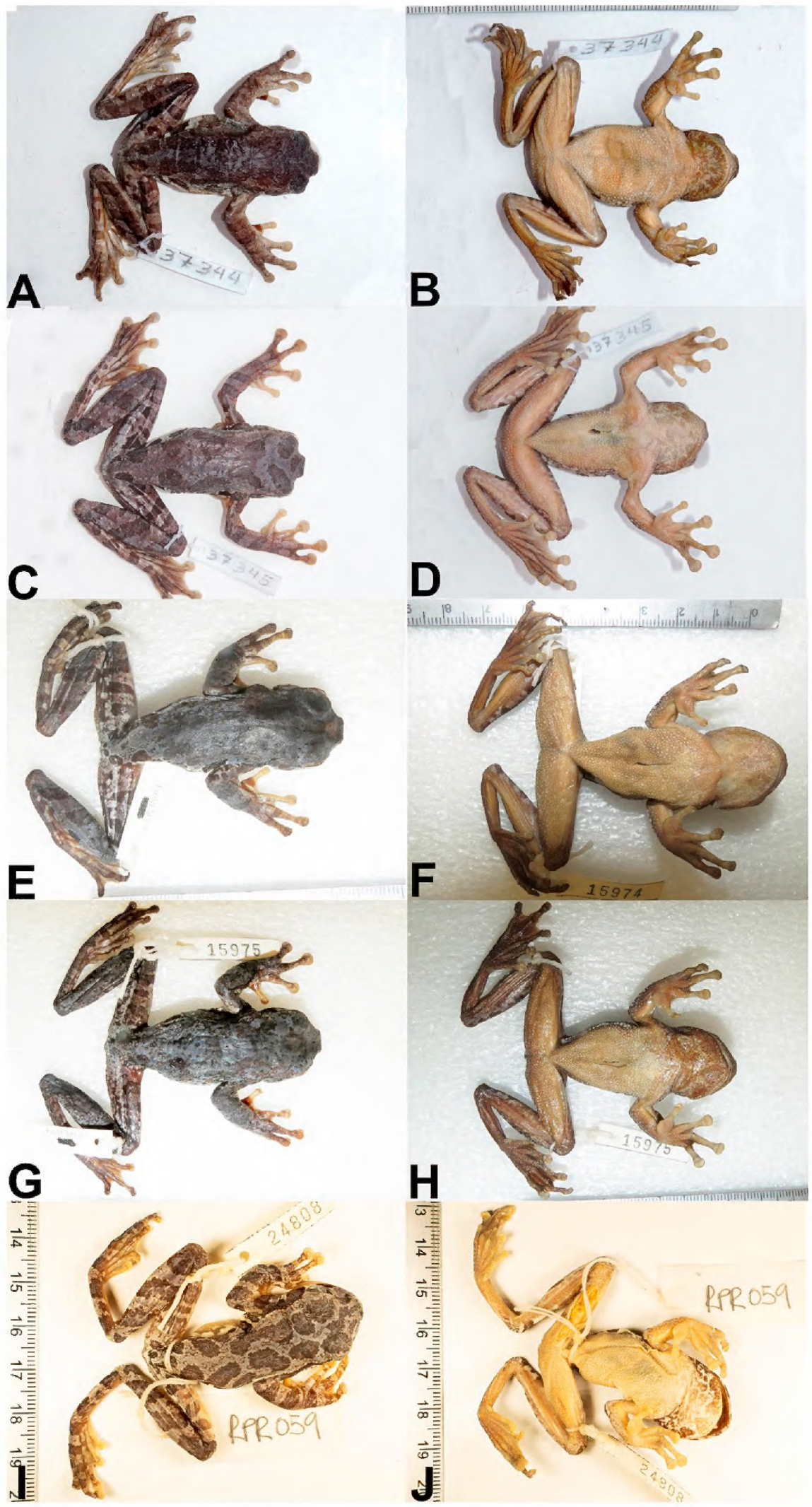

Figure 3. Paratypes of *Charadrahyla viruela* sp. nov. in preservative, showing variation amongst specimens from different localities, all in the Sierra Juárez, Oaxaca in dorsal (**A, C, E, G, I**) and ventral (**B, D, F, H, J**) view.

Santiago Comaltepec (17.6133, -96.3691, 1,715 m asl; datum = WGS84), Oaxaca, Mexico, collected on 19 June 2002 by Adrián Nieto-Montes de Oca; **CNAR 24808**, an adult male, Arroyo Tepetotutla, 6.6 km (airline) S of Santa Cruz Tepetotutla, Municipio de San Felipe Usila (17.6801, -96.5468, 2,030 m asl; datum = WGS84), Oaxaca, collected on 26 April 2009 by Rocío Ponce, Omar Hernández, and Aldo Carrillo; **INIRENA 2949** (CIG 1252), 1 adult female, 4.2 km S of Vista Hermosa, and approx. 1.2 km S of La Esperanza, Municipio de Santiago Comaltepec (17.6133, -96.3691, 1,715 m asl; datum = WGS84), Oaxaca, Mexico, collected on 24 July 2017 by Héctor Franz-Chávez, André J. Grünwald, and Christoph I. Grünwald; **INIRENA 2948** (CIG 2610), 1 adult male, 4.2 km S of Vista Hermosa, and approx. 1.2 km S of La Esperanza, Municipio de Santiago Comaltepec (17.6133, -96.3691, 1,715 m asl; datum = WGS84), Oaxaca, Mexico, collected on 17 August 2025 by Christoph I. Grünwald, Leonardo M. Martínez-Campos, Jonathan D. Wedow, Brandon T. La Forest, Oswaldo Corona-Garduño.

Referred specimens. MEXICO: OAXACA: 8 km S of Yetla, Municipio de San Juan Bautista Valle Nacional (**KU 86967**); 9 km S of Yetla, Municipio de San Juan Bautista Valle Nacional (**KU 64335**); 10.5 km S of Valle Nacional, Municipio de San Juan Bautista Valle Nacional (**KU 136651, KU 136655**); 4.2 km S of Vista Hermosa Municipio de Santiago Comaltepec (**KU 86966**).

MZFC 37344 from approx. 1.2 km S of La Esperanza, Municipio de Santiago Comaltepec, Oaxaca; **MZFC 37345** from Arroyo Tres Manantiales, La Esperanza, Municipio de Santiago Comaltepec, Oaxaca (A, B); **MZFC 15974** from 4.2 km S Vista Hermosa on Hwy 175, ca 1.2 km S La Esperanza, Municipio de Santiago Comaltepec, Oaxaca (C, D); **MZFC 15975** from 4.2 km S Vista Hermosa on Hwy 175, ca 1.2 km S La Esperanza, Municipio de Santiago Comaltepec, Oaxaca (E, F); **CNAR 24808** from Arroyo Tepetotutla, 6.6 km (airline) S of Santa Cruz Tepetotutla, Municipio de San Felipe Usila, Oaxaca (G, H); **INIRENA 2949** from 4.2 km S Vista Hermosa on Hwy 175, ca 1.2 km S La Esperanza, Municipio de Santiago Comaltepec, Oaxaca (I, J).

Diagnosis. A moderately sized *Charadrahyla* (males 53.3–61.8 mm, females to at least 76.8 mm) with a distinctly short snout; extensively tuberculate dorsum; and a unique emerald green dorsal coloration. The dorsal patterning consists of large, dark, military green dorsal blotches, with even darker green outlines and pale green interiors, which in turn are covered by dark green spots on the pale interiors. Some tan blotching is present dorso-laterally. The numerous dorsal tubercles are reddish-brown or tan, colored distinctly from the dorsal ground coloration surrounding them. The overall combination of the dorsal patterning gives the frog a "mossy" appearance. The iris is golden, heavily reticulated with black, giving a "starry-eyed" appearance. This species can be distinguished from all other recognized species of *Charadrahyla* by the combination of the following characters: presence of tuberculate dorsal skin; lack of a rostral keel; lack of axillary membrane; lack of hypertrophied webbing between first and second toe; lack of conical tubercles on either side of cloacal opening; and the distinctively reticulate "starry-eyed" iris. Other identifying characteristics include: snout short in both adult males and females; snout truncate in both dorsal and lateral profiles; nostrils flared, protruding; distinctive white or pale cream blotch on upper lip, located posterior to the eye; subarticular tubercles on some of the fingers distinctly bifid (which fingers varies from specimen to specimen); finger disks pale yellow or cream in coloration, distinctly paler than the pale dorsal coloration of hands; noticeable sexual dimorphism in finger disc size, females with much larger discs; ventral coloration off-white or cream, with no immaculate white spots present on chest; throat coloration cream with large, faded, tan blotches on throat, blotches often squarish in shape; and lateral blotches which are distinct, regular, well defined and highly contrasting compared to other blotches on dorsum. Comparisons between the different species of *Charadrahyla* are given in Table 1.

Comparisons. Phenotypically this species appears to be most similar to *C. nephila* and *C. chaneque*, the former occurring in sympatry or near sympatry at moderate elevations in the Sierra Juárez of northern Oaxaca. *Charadrahyla viruela* differs from *C. nephila* (Comparisons in Fig. 4; characters of *C. nephila* in parenthesis) by possessing tuberculate skin on the entire dorsum (vs. smooth dorsal skin); shorter snout, which is truncate in lateral profile (vs. longer snout, which is rounded in lateral profile); flared nostrils (vs. nostrils not flared); subarticular tubercles on some fingers distinctly bifid (vs. subarticular tubercles rounded); sexual dimorphism in finger-disc size (vs. finger discs same size in both sexes); dorsal ground coloration emerald green, with large dark military green dorsal blotches (vs. dorsal ground coloration brown with large dark brown dorsal blotches); dark dorsal blotches consist of paler green interiors and darker green outlines, and dark green spots on the pale interior (vs. dark brown dorsal blotches which are varyingly sprinkled with green iridophores, and on some individuals iridophores combine to form large areas of green coloration on the large brown dorsal blotches); numerous dorsal tubercles are reddish-brown or tan (vs. dorsal tubercles absent, thus giving dorsal ground coloration a clean look); the overall combination of the dorsal patterning gives the frog a "mossy" appearance (vs. the overall combination of the dorsal patterning gives the frog a "military camouflage" appearance); distinctive white or whitish-yellow blotch on upper lip posterior to eye (vs. no distinctive white blotch on upper lip); iris golden, with heavy black reticulations, giving it a starry appearance (vs. iris solid copper coloration); large tan blotches on throat (vs. brown throat with no dark blotching, occasional white spot present on throat or chest) and lateral blotches which are distinct, regular, well defined and highly contrasting compared to other blotches on dorsum (vs. highly contrasting lateral blotches absent).

Charadrahyla viruela differs from *C. chaneque* as follows: always possessing tuberculate dorsal skin (vs. dorsal skin smooth or slightly tuberculate); dorsal tubercles reddish brown (vs. dorsal tubercles yellow, tan or brown but never reddish); dorsal ground coloration emerald green, with large dark military green dorsal blotches (vs. dorsal ground coloration tan to pale brown, with larger dark brown dorsal blotches); dark green dorsal blotches consist of paler green interiors and darker green outlines, and dark green spots on the pale interior (vs. dark dorsal blotches which for the most part lack green iridophores); numerous dorsal tubercles are reddish-brown or tan (vs. dorsal tubercles same color as skin around them, not reddish); green iridophores not widespread on dorsal ground coloration (vs. green iridophores widespread on dorsal ground coloration, giving some specimens a greenish appearance); the overall combination of the dorsal coloration and patterning gives the frog a "mossy" appearance (vs. overall dorsal coloration pale with dark blotches, much cleaner than in *C. viruela*); snout truncate in lateral profile (vs. bluntly rounded); subarticular tubercles on some fingers distinctly bifid (vs. large and rounded, none bifid). Comparisons between *C. viruela*, *C. nephila* and *C. chaneque* are given in Table 2.

Table 1. Variation and similarities of selected morphological characters amongst related species of *Charadrahyla*.

Species	*Charadrahyla viruela* **sp. nov.**	*Charadrahyla nephila*	*Charadrahyla chaneque*	*Charadrahyla sakbah*	*Charadrahyla esperancensis*	*Charadrahyla altipotens*	*Charadrahyla trux*	*Charadrahyla tecuani*	*Charadrahyla taeniopus*	*Charadrahyla juanitae*	*Charadrahyla pinorum*
Adult Size ♂	53.3–61.8	53.7–61.9	56.0–60.7	67.91–73.21	48.9–54.9	69.4–80.6	79.1–81.0	52.5–57.8	48.1–65.9	27.7–35.6	28.5–33.1
Adult Size ♀	64.7–73.2	70.0–79.3	79.3	81.15–85.75	none known	77.7–78.8	unknown	unknown	65.0–70	35.0–39.8	30.0–34.6
Dorsal Skin Condition	Tuberculate	Smooth	Variable, when present tubercles not red tipped	Smooth	Smooth	Smooth	Smooth	Smooth	Smooth	Smooth	Smooth
Snout Length	Snout short, in both sexes	Snout short, in both sexes	Snout short in both sexes	Snout moderately long	Snout long	Snout long	Snout moderately Long	Snout moderately long	Snout long in ♂, moderately long in ♀	Snout short	Snout moderately long
Snout Shape Dorsal Profile	Snout truncate in dorsal profile	Rounded	Rounded	Rounded	Pointed, in dorsal profile	Acuminate, not truncate	Moderately accuminate	Moderately accuminate	Acuminate in ♂, truncate in ♀	Rounded	Bluntly rounded
Snout Shape Lateral Profile	Snout truncated in lateral profile	Rounded	Slightly rounded in lateral profile	Rounded or acuminate, NOT truncate	Protruding in lateral profile	Acutely rounded, not truncate	Rounded, not truncate	Rounded, not truncate	Protruding in ♂, rounded in ♀	Rounded	Rounded
Nostrils Flared?	Flared nostrils	Slightly flared	Slightly flared	Slightly flared	Flared nostrils	Slightly flared	Slightly flared	Slightly flared	Flared nostrils	Slightly flared	Flared nostrils
Rostral Keel?	No	No	No	No	Distinct rostral keel	Moderate keel	No	No	No	No	No
Axillary Membrane?	Absent	Absent	Absent	Present	Absent	Present	Absent	Absent	Absent	Present	Present
Webbing between 1st & 2nd toe hypertrophied?	No	No	No	Yes, hypertrophied	No	No	Yes, hypertrophied	Yes, hypertrophied	No	No	No
Finger Disk Coloration	Finger disks pale coloration, distinctly paler than dorsal coloration of hands.	Finger disks pales, same color as pale bands on hands	Same color as pale bands on hands	Variable, usually same color as hands, not paler	Same color as hands	Same coloration as anterior parts of fingers	Much paler than dorsal coloration of hand	Same color as pale bands on hands	Variable, but usually same color as pale portions of forearms	Same coloration as fingers	Same coloration as fingers
Subarticular Tubercle Condition on Some Fingers	Bifid on some fingers	Not bifid	Not bifid	Not bifid	Not bifid	Round, not bifid	Round, not bifid	Round, not bifid	Round, not bifid	Subconical, barely bifid	Subconical, barely bifid
Iris Pattern	Iris golden, heavily reticulated with black, giving a "starry-eyed" appearance	Iris copper-brown, solid-colored, not reticulated and no "starry-eyed" appearance	Iris copper-brown, solid-colored, not reticulated and no "starry -eyed" appearance	bronze, with fine dark lines, not reticulations and thus no "starry-eyed" appearance	Solid brown coloration with no dark patterning	Iris pale bronze colored, with fine dark reticulation, not "starry-eyed"	Iris golden with fine dark lines, not "starry-eyed"	Iris brown with fine dark lines, not "starry-eyed"	Iris color variable between juveniles and adults; adults bronze with copper periphery, not "starry-eyed"	Iris pale copper with fine black lines, not "starry-eyed"	Iris pale copper with fine black lines, not "starry-eyed"

Species	*Charadrahyla viruela* sp. nov.	*Charadrahyla nephila*	*Charadrahyla chaneque*	*Charadrahyla sakbah*	*Charadrahyla esperancensis*	*Charadrahyla altipotens*	*Charadrahyla trux*	*Charadrahyla tecuani*	*Charadrahyla taeniopus*	*Charadrahyla juanitae*	*Charadrahyla pinorum*
Presence of Distinctive Labial Blotch Present	Distinctive white blotch present on upper lip, posterior to the eye	No distinctive white blotch on lip, posterior upper lip pale	No distinctive white blotch on lip, posterior upper lip pale	Variable, but not with distinctive white spot	Upper lip unmarked	Variable, unmarked, no white blotch	No distinct pale blotch on upper lip	No distinct pale blotch on upper lip	No distinct pale blotch on upper lip	No distinct pale blotch on lip	Distinctive white blotch on lip, from middle of eye to beneath tympanum
Dorsal Ground Coloration	Dorsal ground coloration emerald green, with dark brown blotches covered in emerald green iridophores, dorsal tubercles reddish-brown, overall "mossy" appearance	Dorsal coloration light brown, with large dark brown blotches, and variable green highlights, dorsal tubercles absent, overall appearance "military camouflage"	Dorsal ground coloration light brown mixed with green iridophores, with dark brown blotches, dorsal tubercles same color as adjacent skin	Variable, usually brown dorsal coloration with darker brown blotches, green may be present, no dorsal tubercles and no "mossy appearance"	Solid brown coloration with some dark spots, no dark blotches and no dorsal tubercles, no "mossy appearance"	Usually pale green or brown, with darker green blotches, no large dark brown blotches and no dorsal tubercles, no "mossy appearance"	Brown, with darker brown blotches and barring on legs and scattered clusters of green iridophores	Brown, with darker brown blotches and barring on legs and scattered clusters of green iridophores	Dorsal coloration extremely variable, sometimes green, but never with combination of colors creating overall "mossy" appearance	Dorsal coloration pale brown with darker brown markings, no "mossy" appearance	Dorsal coloration pale tan with pale brown irregular markings, no "mossy" appearance
Ventral Coloration	Pale cream ventral coloration on throat, with large darker brown blotches or reticulations	Dark brown ventral coloration on throat, with pale white, yellow or cream dots or stippling; no dark blotches	Ventral coloration pale cream, In males, pale cream ventral coloration on throat, with small dark brown blotches, in females throat dark	Pale cream ventral coloration with no dark markings	Dark reticulation, not large squarish blotches	Creamy yellow, unmarked, throat unmarked	Dull creamy brown with sparse reticulation throat	Pale yellow venter with a few dark brown markings on throat, no large blotches	Variable, usually pale with variable brown coloration on throat, not large blotches	Venter white, throat white with, with minor dark speckling, no blotches	Venter creamy white, throat pale, without dark markings
Lateral Blotch Condition	Three to four large, rectangular or sub-rectangular distinct, dark lateral blotches, outlined in white, located on the posterior portion of the flank	Numerous small, rounded lateral blotches, not darker or more distinct than other dorsal blotches and not outlined in white	Posterior flanks have some small, dark brown, blotches, usually not outlined white; even when outlined, not distinct and contrasting	Dark blotches variable, some specimens do have distinct high-contrast blotches	Distinctive brown and yellow reticulations, no rectangular blotches on flanks	Bright creamy yellow with dark brown reticulations and spots, no large lateral blotches	Distinctive brown and yellow reticulations, no rectangular blotches on flanks	Yellow with numerous small brown blotches, not rectangular and not large as in *C. viruela*	Variable, distinct lateral blotches may be present but not large and squarish	No distinct, high contrast, dorso-lateral blotches	No distinct, high contrast, dorso-lateral blotches
Cloacal tubercles present?	Present	Present, indistinct	Present	Present	Absent	Absent	Present	Present, large	Absent	Present	Present, very large!
Nuptial excrescences	Present	Present	Present	Present	Present	Absent	Present	Present	Present	Present	Absent

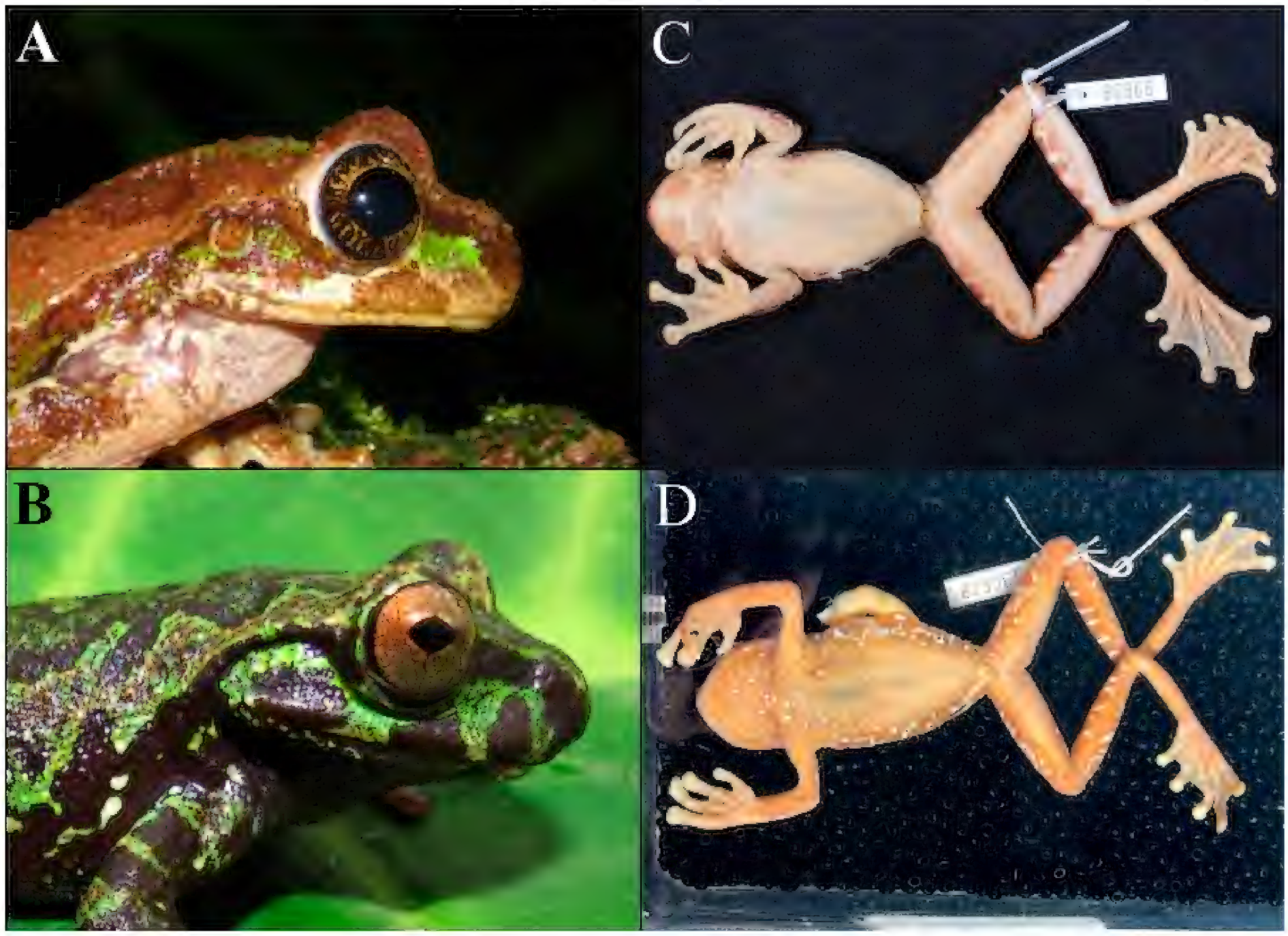

Figure 4. Comparative characters between *Charadrahyla viruela* sp. nov. and *C. nephila*. **A.** Lateral view of head of a live individual of *C. viruela* from ca 1.2 km S of La Esperanza, Municipio de Santiago Comaltepec, Oaxaca, which shows the truncate snout in lateral profile; the "starry-eyed" reticulated iris; and the characteristic white blotch on the lip posterior to the eyes; **B.** Lateral view of head of a live individual of *C. nephila* from 6.6 km S of La Esperanza, at Arroyo Buena Vista, Municipio de Santiago Comaltepec, Oaxaca which shows the characteristically rounded snout in lateral profile; the solid, copper-colored iris with no reticulations; and the lack of a distinctive white blotch on the upper lip posterior to the eye; **C.** Ventral view of a preserved *C. viruela* (KU 86966) from 4.2 km S of Vista Hermosa, Municipio de Santiago Comaltepec, Oaxaca which shows the characteristic pale cream to whitish ventral coloration; the seven large tan blotches on the throat; the lack of immaculate white spotting on the throat and chest; and the dark brown banding instead of white striping on ventrolateral portions of the thighs and tibia; **D.** Ventral view of a preserved *C. nephila* (136678) from 11.9 km S of Vista Hermosa, Municipio de Santiago Comaltepec, Oaxaca which shows the characteristic darker tan to brown ventral coloration, lack of tan blotches on throat, presence of immaculate white spotting on throat, chest, and upper abdomen, and immaculate white striping on the ventrolateral portions of the thighs and tibia.

Charadrahyla viruela can be distinguished from the sympatric *C. esperancensis* by its tuberculate skin (vs. smooth), much shorter snout, truncate from a lateral profile (vs. longer snout, protruding in lateral profile), lack of a rostral keel (vs. distinct rostral keel present), finger discs pale yellowish, brighter than pale coloration on hands (vs. finger discs same color as pale portions of hands), iris golden with black reticulation, giving starry-eyed appearance (vs. iris solid brown with no dark patterning), presence of large regular blotches on the lateral flanks and lack of yellow reticulation (vs. distinctive brown and yellow reticulations, no rectangular blotches on dorsolateral surfaces), distinct white blotch up upper lip posterior to eye (vs. upper lip unmarked, with no distinctive pale blotches); unique emerald green dorsal coloration with dark green dorsal blotches, extensive emerald green iridophores, dark green spotting on iridophores, and reddish-brown dorsal tubercles, the combination of which gives the frog an overall "mossy appearance" (vs. dorsal coloration solid brown coloration with some darker brown spots); and by the presence large, dark, squarish blotches present on throat (vs. dark reticulations present on throat, with no large blotches).

The widespread *Charadrahyla taeniopus* is a highly variable species, and some of its pattern classes present a similar dorsal coloration with an overall "mossy" appearance as in *C. viruela*. However, *C. viruela* can be distinguished from *C. taeniopus* by being a much smaller frog, with adult males 53.3–61.8 mm (vs. up to 65.9); with tuberculate dorsal skin (vs. smooth); short, truncate snout in both adult male and female lateral profile (vs. rounded snout in lateral profile in females and subadults,

Table 2. Comparison of diagnostic characters of *Charadrahyla viruela* sp. nov., *Charadrahyla nephila* and *Charadrahyla chaneque*.

***Charadrahyla viruela* sp. nov.**	***Charadrahyla nephila***	***Charadrahyla chaneque***
Tuberculate dorsal skin, with tubercles reddish or reddish-brown in coloration, distinct from dorsal ground coloration.	Smooth dorsal skin, no tubercles.	Variable, dorsal may or may not present tubercles. When present tubercles, same color as dorsal ground coloration.
Snout truncate in both dorsal and lateral profiles.	Snout rounded, in both dorsal and lateral profiles.	Snout rounded in dorsal profile, rounded or slightly truncated in lateral profile.
Subarticular tubercles on some fingers are bifid.	Subarticular tubercles on fingers not bifid.	Subarticular tubercles on fingers not bifid.
Dorsal coloration emerald green with dark green blotches, emerald green iridophores and reddish-brown dorsal tubercles, the combination of which gives overall "mossy" appearance.	Dorsal coloration light brown, with large dark brown blotches, with green iridophores that combine to form large areas of green coloration large brown dorsal blotches; dorsal tubercles absent, the overall combination of the dorsal patterning gives the frog a "military camouflage" appearance.	Dorsal ground coloration tan to pale brown, with larger dark brown dorsal blotches, which for the most part lack green iridophores; green iridophores widespread on dorsal ground coloration, giving some specimens a greenish appearance; dorsal tubercles present, same color as skin around them.
Three to four large, rectangular or sub-rectangular distinct, dark lateral blotches, outlined in white, located on the posterior portion of the flank.	Numerous small, rounded lateral blotches, not darker or more distinct than other dorsal blotches and not outlined in white.	Posterial flanks have some small, dark brown, blotches, usually not outlined white. Even when outlined, not distinct and contrasting.
Distinctive white blotch present on upper lip posterior to eye.	No distinctive white blotch present on upper lip posterior to eye.	No distinctive white blotch present on upper lip posterior to eye.
Pale cream ventral coloration on throat, with large darker brown blotches or reticulations.	Dark brown ventral coloration on throat, with pale white, yellow or cream dots or stippling. No dark blotches.	Males with pale cream ventral coloration on throat and small darker brown blotches, females with dark brown dorsal coloration on throat.
Finger disks distinctly paler than the pale dorsal coloration of hands.	Finger disk coloration variable, distinctly paler than the pale dorsal coloration of hands.	Finger disks have the same coloration as dorsal coloration of hands.
Iris golden, heavily reticulated with black, giving a distinctive "starry-eyed" appearance.	Iris bronze or copper, solid colored and no "starry-eyed" appearance.	Iris bronze or copper, solid colored and no "starry-eyed" appearance.

elongated and acuminate in adult males); subarticular tubercles on some fingers bifid (vs. rounded or conical, not bifid); presence of the distinct white blotch on the upper lip (vs. no distinctive white blotch on lip); and the presence of cloacal tubercles (vs. absent).

From the four large species of *Charadrahyla* from the Sierra Madre del Sur (*C. altipotens*, *C. tecuani*, *C. trux*, and *C. sakbah*), *C. viruela* can be easily distinguished by its much smaller adult size not exceeding 61.8 mm in males (vs. larger than 55.3 mm, *C. tecuani* being the smallest of the four, usually males above 60.0 mm); tuberculate dorsal skin (vs. smooth, and slightly tuberculate in *C. tecuani*); short, truncate snout (vs. longer snout, slightly protruding in males); iris golden with black reticulation, with starry appearance (vs. iris with thin black lines, no reticulations and not "starry" in appearance); distinct white spot on upper lip posterior to eye (vs. no distinct white spot on lip); unique emerald green dorsal coloration with dark green dorsal blotches, extensive emerald green iridophores, dark green spotting on iridophores, and reddish-brown dorsal tubercles, the combination of which gives the frog an overall "mossy appearance" (vs. dorsal coloration variable, but not emerald green with an overall mossy appearance); it furthermore can be distinguished from *C. sakbah*, *C. tecuani*, and *C. trux* by lack of hypertrophied tissue between the first and second toes (vs. present in males of those species).

Finally, *C. viruela* differs from the two smaller members of the genus (*C. pinorum* and *C. juanitae*) by presenting an external tympanum (vs. lacking external tympanum); its larger size of males 53.3–61.8 mm (vs. 27.7–35.8 mm); distinctly tuberculate dorsum (vs. smooth dorsum); lack of axillary membrane (vs. presence of axillary membrane, albeit small); pale yellow finger discs on hands which are brighter than pale portions of hands (vs. finger disks same color as pale portions of hands); iris golden with dark reticulations, giving starry-eyed appearance (vs. iris pale brown with thin black lines, no starry-eyed appearance); unique emerald green dorsal coloration with dark green dorsal blotches, extensive emerald green iridophores, dark green spotting on iridophores, and reddish-brown dorsal tubercles, the combination of which gives the frog an overall "mossy appearance" (vs. dorsal coloration pale brown with darker brown blotches, not emerald green with an overall "mossy" appearance); throat with large, squarish, brown blotches (vs. throat pale, unmarked, with no dark blotches); presence of distinct, highly contrasting dorsolateral blotches (vs. no distinct, high contrast dorsolateral blotches); cloacal tubercles indistinct (distinctive cloacal tubercles present both above and below cloacal opening). It is noteworthy that these two smaller species are the only species within *Charadrahyla* that share bifid subarticular tubercles (on the third and fourth fingers) and *C. pinorum* is the only other species of the genus with a characteristic, distinct, white spot on the upper lip posterior to the eye.

Description of the holotype. Subadult female, relatively large (51.5 mm SVL); head slightly wider (18.3 mm) than long (17.8 mm), top of head flat; head as wide as body; HL/SVL ratio 0.34; nostrils protuberant, sub-oval, directed diagonally; snout truncate in dorsal profile with no distinct dorsal rostral keel; snout short and truncate in both dorsal and lateral profiles, slightly sub-rounded in lateral profile; tympanum indistinct, round, small, greatest

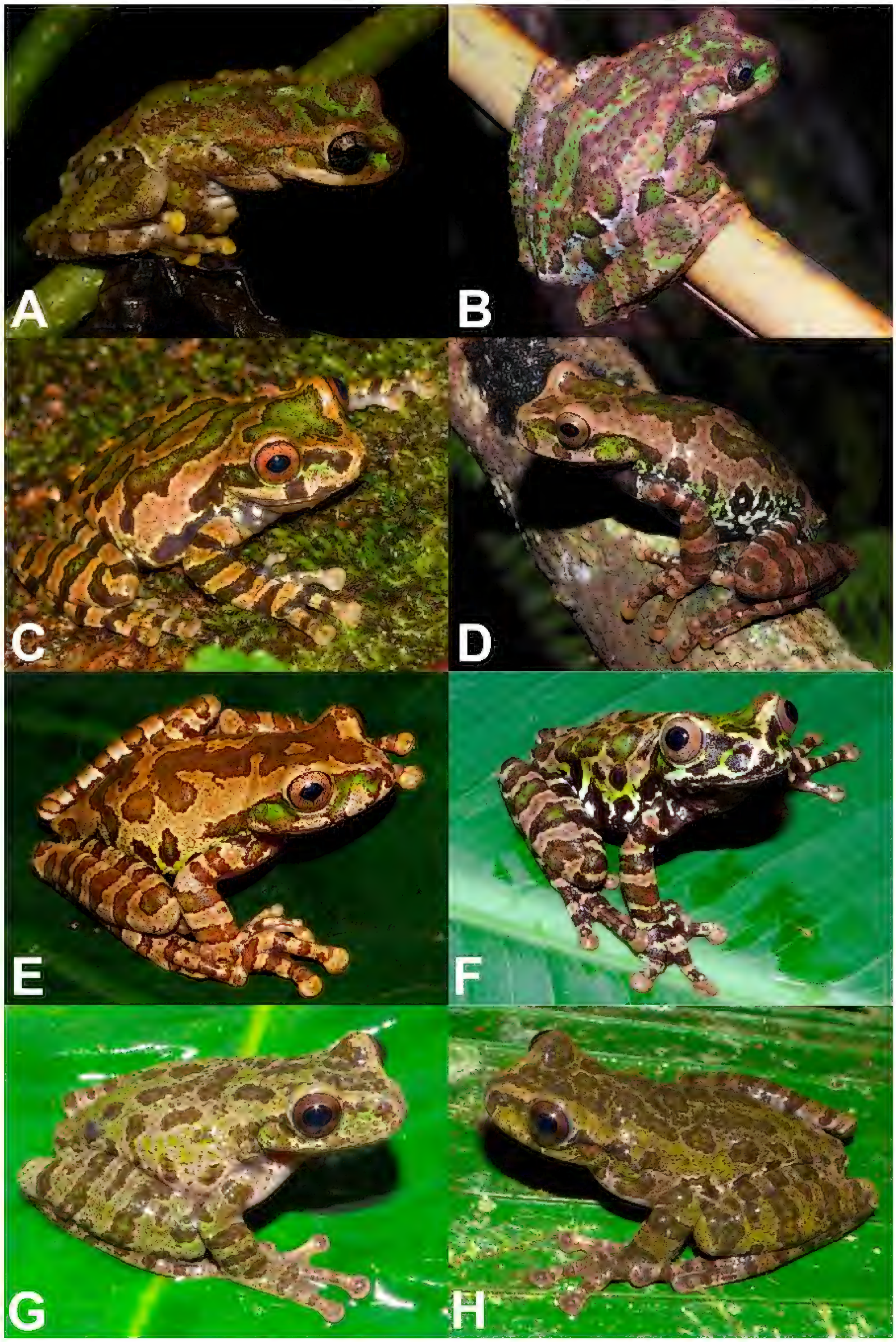

Figure 5. Comparison of individuals of *Charadrahyla* species. We have included photos of live specimens whenever possible, but were unable to obtain photos of live individuals of two populations. **A.** *Charadrahyla viruela* sp. nov. (**MZFC 37345**, paratype) from Sierra Juárez, at Arroyo Tres Manantiales, La Esperanza, Municipio de Santiago Comaltepec, Oaxaca, at 1,688 m asl; **B.** *Charadrahyla viruela* sp. nov. from Sierra Juárez, at La Esperanza, Municipio de Santiago Comaltepec, Oaxaca, at 1,630 m asl, photo by Jesse Delia; **C.** *Charadrahyla nephila* from Sierra Juárez, in the upper reaches of Arroyo Tepetotutla, Municipio de San Felipe Usila, Oaxaca, at 2,600 m asl, photo by Rogelio Simón Salvador; **D.** *Charadrahyla nephila* Arroyo Grande at 16 km S of La Esperanza, Municipio de Santiago Comaltepec, Oaxaca, at 2,150 m asl (**CIG 1871**); **E, F.** *Charadrahyla nephila* type-locality, 5.8 km SW of Totontepec, Municipio de Totontepec Villa de Morelos, Oaxaca, at 2,000 m asl; **G, H.** *Charadrahyla chaneque* type-locality, 6.2 km S of Rayón Mescalapa, Municipio de Rayón, Chiapas at 1,790 m asl.

width of tympanum 2.3 mm; distinct supratympanic fold from eye, above tympanum and down to above arm insertion, barely covering upper edge of tympanum; canthus moderately angular; loreal region concave; lips moderate and not flared; greatest diameter of eye 5.7 mm; tympanum width to eye-diameter 0.40; eyelid width 4.3 mm, approximately 40% of the IOD; HaL 19.1 mm; fingers long, 3FL 12.8 mm, 4FL 9.7 mm, 3FL/SVL ratio 0.24; finger lengths 1 < 4 < 2 < 3; fingers moderately webbed, hand webbing formula: **I** 2–2½ **II** 1½–2 **III** 1¾–2 **IV**; finger discs large, 3FPW 3.2 mm, 3FPW/3FW ratio 2.7; 4FPW 3.1 mm, 4FPW/4FW ratio 1.8, 4FPW/TW ratio 2.3; subarticular tubercles on fingers large, tubercle on first finger oval, slightly bifid, tubercle on second finger rounded, and tubercles on third and fourth fingers bifid; indistinct supernumerary pustules rounded; no distinct palmar tubercle, rather numerous small tubercles present on palm; no dermal fold on wrist; enlarged tubercles on ventrolateral edge of forearm in row, interrupted, not forming a continuous fold, 5 tubercles on left arm and 6 on right arm; there is no axillary membrane; FL 23.8, toes long, 3TL 16.8 mm, 4TL 20.1 mm, 4TL/SVL ratio 0.39; toe lengths 1 < 2 < 5 < 3 < 4; toes fully webbed, webbing not hyper-trophied, foot webbing formula: **I** 1-–1 **II** 1–1 **III** 1–1½ **IV** 1½–2 **V**; toe discs large, 4TPW 2.8 mm, 4TPW/4TW ratio 2.15; toe subarticular tubercles moderately large, round, not bifid: indistinct supernumerary pustules rounded; inner metatarsal tubercle distinct, oval; outer metatarsal tubercle indistinct, not noticeably larger than surrounding tubercles; tarsal fold distinct; a row of slightly raised tubercles on tarsus; tibio-tarsal articulation extends to past snout; cloacal opening directed posteroventrally at mid-level of thighs; cloacal sheath moderately long, grooved medially; no transverse fold above cloacal opening; no enlarged tubercles around vent. Skin on dorsum distinctly tuberculate; lateral skin smooth but slightly shagreen; skin on throat smooth, ventral skin of body and upper thighs granular; ventral skin on arms, lower thighs, tibia and tarsus smooth. The tongue is cordiform, notched posteriorly, and barely free posteriorly. Dentigerous processes of prevomers six on each process; large and posteromedially inclined between two moderate-sized ovoid choanae. Pupil horizontal. We have included illustrations of the ventral surface of the hand (Fig. 6A) and the foot (Fig. 6B) of the holotype.

In life, holotype with emerald-green dorsal coloration on dorsal surfaces of the head back, arms, and legs. Numerous dark green blotches were present on the head and back, these blotches a darker green than the emerald-green ground coloration and outlined in dark brown. Numerous dorsal tubercles were reddish-brown when present on the emerald-green dorsal ground coloration, or dark brown when present on the dark green dorsal blotches. This dorsal coloration creates an overall mossy-like appearance. Top of head emerald green, with numerous dark green blotches outlined in dark brown, and tubercles reddish brown. A continuous line of reddish-brown coloration stretches from the naris, along the canthus rostralis, continues posteriorly along the eyelids and along the upper half of the supratympanic fold, where it fades to pale tan with less reddish undertones. No interorbital bar present. Anterior portion of lip with reddish brown ground coloration and dark brown spots that have a dark green center. Lip posterior to eye with characteristic white blotch, paler than whitish ground coloration on flanks. Tympanum brown, lower portion of supratympanic fold

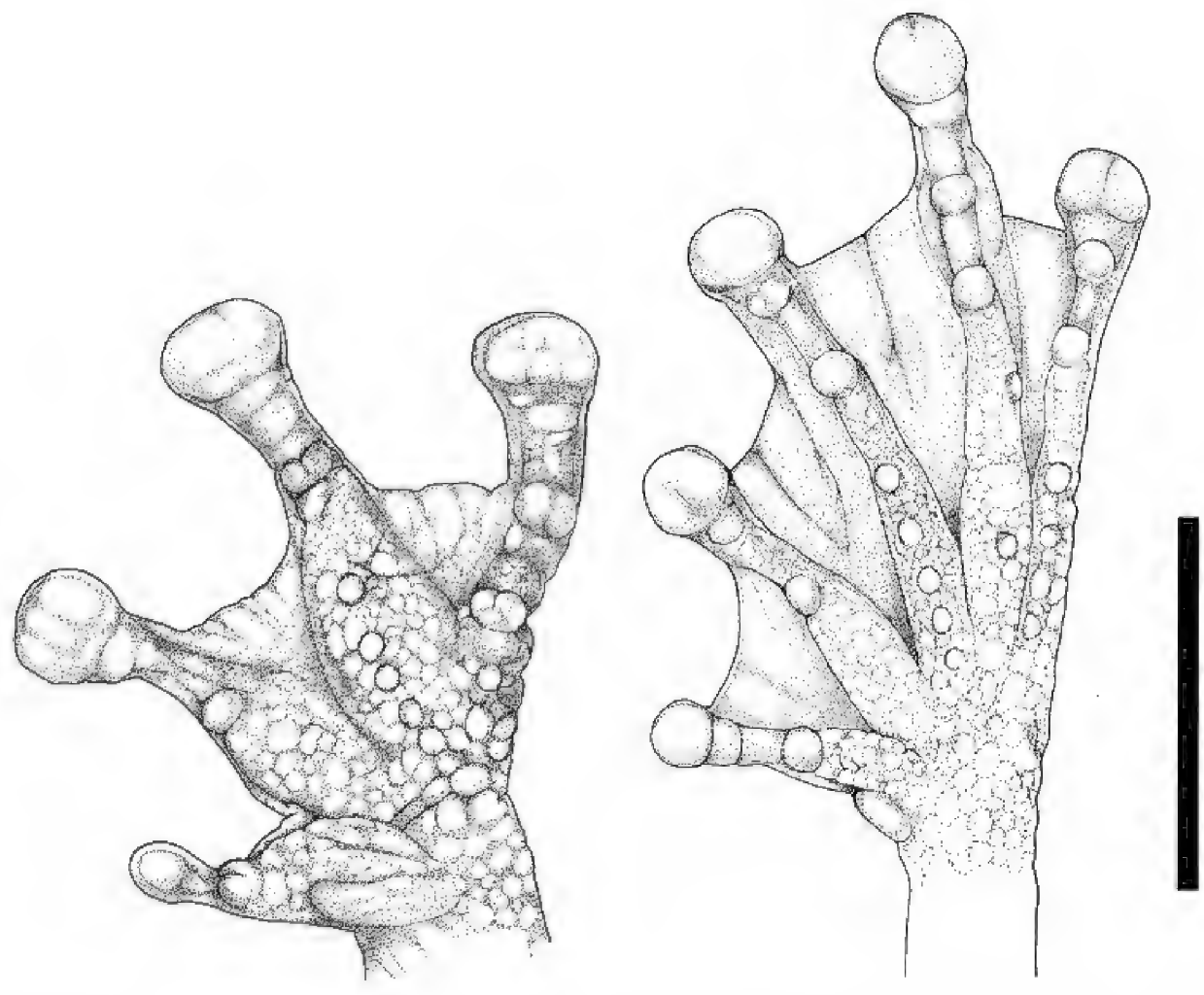

Figure 6. Ventral aspect of hand **A.** and foot **B.** of *Charadrahyla viruela* sp. nov. INIRENA 2950, Highway 175 between Vista Hermosa and La Esperanza, 1.9 km ENE of La Esperanza, Municipio de Santiago Comaltepec, Oaxaca, Mexico. The black bar represents 10 mm. Illustrations by Iván Ahumada-Carrillo.

dark brown. Iris golden with black reticulations which gave it a characteristic "starry-eyed" appearance. Flanks whitish, 5–6 dark brown blotches with dark green centers. Hands and feet brown with darker brown banding, finger discs were a characteristic pale-yellow coloration in life. Throat and ventral surfaces of abdomen whitish cream, with tan blotches on the throat. Ventral colorations of arms, legs, hands and feet whitish.

Coloration in preservative is pale brown on dorsal surfaces of head, body, arms, and legs, slightly darker than body, whereas arms and legs slightly paler, and reddish, than body. Dark brown blotches present on dorsum and flanks, dark brown banding on arms, legs, hands, feet, fingers, and toes. Dark brown dorsal blotches and barring not outlined in black. Dorsal surface of flanks pale cream to whitish ventrolaterally, with dark brown blotches. Distinctive pale blotch on upper lip posterior to eye whitish. Pale tan line from naris posterior to eyelids and upper supratympanic fold, lower supratympanic fold dark brown. Ventral surfaces of body uniform cream colored and unmarked, except on the throat, which is cream but covered by 10–11 large tan blotches. Ventral surfaces of arms, legs, hands, and feet cream and unmarked. Iris milky grey, pupil transparent (Figs 2B–C).

Measurements of the holotype (in millimeters): SVL 51.5, HL 17.8, HW 18.3, ED 5.7, ELW 4.3, IOD 10.7, TW 2.3, FeL 24.4, TiL 28.2, TaL 17.7, FL 23.8, HaL 19.1, FaL 15.1, UaL 10.0, END 4.3, IND 6.5, ETD 2.0, 1FL 7.5, 2FL 10.2, 3FPW 3.2, 3FW 1.2, 3FL 12.8, 4FPW 3.1, 4FW 1.7, 4FL 9.7, 1TL 6.9, 2TL 9.2, 3TL 16.8, 4TPW 2.8, 4TW 1.3, 4TL 20.1, 5TL 14.5.

Ratios of characters. HL/SVL 0.35, HW/SVL 0.36, HL/HW 0.97, IOD/HW 0.58, ED/HW 0.31, TW/ED 0.40, TW/ETD 1.15, 3FPW/3FW 2.67, 4FPW/4FW 1.82, 4TPW/4TW 2.15, HaL/SVL 0.37, TiL/SVL 0.55, FL/SVL 0.46.

Variation. Morphological variation of *C. viruela* is presented in Table 3. *Charadrahyla viruela* does not appear to exhibit any sexual dimorphism in snout shape, or other proportions, other than that females seem to reach a larger size than males (76.8 mm vs. 53.5–61.8 mm SVL, respectively). The one sexual dimorphic proportion that we were able to find is that females appear to have larger finger discs than males, with third finger pad width 2.4–2.7 times the width of finger in females (vs. 1.7–2.3 times the width of the finger in males); and the fourth finger pad width 1.8–2.2 times the width of the finger in females (vs. 1.1–2.0; times the width of the finger in males). One adult male (MZFC 37344) has nuptial excrescences, composed of fine, brown spicules on an ossified, enlarged prepollex and vocal slits. The intensity of the dark blotches on the throat varies broadly in life, most individuals in life the tan blotches are readily discernable. These blotches fade after preservation, and may appear to be absent in some preserved specimen (MZFC 15974; KU 136655). The shape of subarticular tubercles varies from specimen to specimen, for example MZFC 37344 on both hands the subarticular tubercles on fingers 3 and 4 are bifid, the rest rounded; on MZFC 37345 on the left hand the subarticular tubercle on the third finger is slightly bifid, the rest rounded, but on right hand subarticular tubercles of fingers 2 and 3 are distinctly bifid.

Advertisement call. The advertisement call of the males of this species has not been recorded and thus is not described here. However, the species does call, as one individual was collected and photographed while calling from a tree, 2.5 m above the stream 1.2 km S of La Esperanza, Oaxaca on 17 August 2025 (CIG 2610). The call was described as similar to *C. chaneque* and *C. nephila* (B. La Forest, pers. comm.)

Distribution and ecology. *Charadrahyla viruela* appears to be endemic to moderate elevations on the northern slopes of the Sierra Juárez of northern Oaxaca (Fig. 7). This frog occurs at elevations between 600–1,725 m asl and has been collected in lower montane rainforest and lower montane cloud forest. There are at least three assemblages of cloud forest in the Sierra Juárez, including a lower montane cloud forest, an upper montane cloud forest and a cold high-elevation cloud forest. We have included a photo of the type-locality as an example of what the cloud forest looks like within the range (Fig. 8).

Etymology. From latin, *viruela*, meaning small pox, in reference to the extensive tubercles on the dorsum of this species.

Remarks. Mendelson and Campbell (1999) designated 53 specimens as paratypes of *C. nephila* and referred 35 more specimens to that species. We examined photographs of 51 of the paratypes of *C. nephila* (KU 136660 and KU 136687 are not locatable at the collection and may be missing; A Motta & G. Weatherman, pers. comm.), and we were able to refer two of these paratypes to *C. viruela* (KU 136651, 136655) and positively identify 40 paratypes as *C. nephila* (see Appendix 1). The remaining nine paratypes which were examined were not positively identifiable in photographs, either because they are juveniles, badly preserved, or show intermediate pattern in dorsal coloration; and unfortunately, no photographs are available showing the paratypes in life. It should be noted that most unidentifiable specimens originate from the locality listed as "4.2 km S of Vista Hermosa". Both *C. nephila* and *C. viruela* have been reported from the locality "4.2 km S of Vista Hermosa". Interestingly, J. Caldwell (pers. notes) reported this locality to be at 1,580 m asl and all the specimens collected by her at this locality are *C. viruela*, whereas W. Duellman (pers. notes) reports this locality to be at 1,800 m asl and the specimens collected by him are *C. nephila*. We have only collected *C. viruela* at this locality, and propose one of two scenarios may explain the confusion around this locality. It is possible that two different streams are referenced with this locality, and the confusion may stem from Caldwell referencing the Mestizo lumber camp as "Vista Hermosa" (17.6321, -96.3424°) and Duellman referencing the Chinanteca village of La Esperanza as "Vista Hermosa" (17.6285, -96.3675°). The other scenario is that both species occur at the same locality (parking spot), along the same stream, but one occurs at elevations below Highway 175 down towards the town, and the other occurs above Highway 175 up towards the summit. Different collectors may have decided to take different directions (down-

Table 3. Mensural variation of morphometrics of *Charadrahyla viruela* sp. nov., Abbreviations used are as follows: SVL (snout–vent length); HL(head length); HW (head width); ED (diameter of eye); ELW (eyelid width); IOD (interorbital distance); TW (tympanum width); END (eye–naris distance); IND (internarial distance); ETD (eye–tympanum distance); FeL (femur length); TiL (tibia length); TaL (tarsus length); FL (foot length); HaL (hand length); FaL (forearm length); UaL (upper arm length); 1FL (1st finger length); 2FL (2nd finger length); 3FPW (width of pad on 3rd finger); 3FW (width of 3rd finger); 3FL (length of 3rd finger); 4FPW (width of pad on 4th finger; 4FW (width of 4th finger); 4FL (length of 4th finger); 1TL (length of 1st toe); length of 2nd toe (2TL); 3TL (length of 3rd toe); 4TPW (width of pad on 4th toe); 4TW (width of 4th toe); 4TL (length of 4th toe); 5TL length of 5th toe. All measurements in mm.

Species	***Charadrahyla viruela* sp. nov.**							
Sp. #	**INIRENA 2950**	**MZFC 37344**	**MZFC 37345**	**MZFC 15974**	**MZFC 15975**	**CNAR 24808**	**INIRENA 2949**	**INIRENA 2948**
Sex	**FEMALE**	**MALE**	**FEMALE**	**MALE**	**MALE**	**MALE**	**FEMALE**	**MALE**
SVL	51.5	54.7	66.9	54.8	54.8	49.0	76.8	61.8
HL	17.8	16.3	19.5	18.6	17.6	17.2	23.3	17.9
HW	18.3	18.7	23.5	18.8	18.0	17.6	24.2	19.3
HL/SVL	0.3	0.3	0.3	0.3	0.3	0.4	0.3	0.3
HL/HW	1.0	0.9	0.8	1.0	1.0	1.0	1.0	0.9
ED	5.7	5.8	7.6	6.0	5.8	5.6	7.6	6.3
ED/SVL	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1
ED/HL	0.3	0.4	0.4	0.3	0.3	0.3	0.3	0.3
ELW	4.3	5.8	7.5	6.9	4.6	5.5	6.0	4.9
IOD	5.0	5.9	7.7	3.9	4.4	5.2	9.0	7.1
TW	2.3	2.2	2.7	2.6	2.5	2.3	3.3	4.0
TW/ED	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.4
END	4.3	3.7	5.0	4.3	4.1	3.8	5.8	4.4
END/HL	0.2	0.2	0.3	0.2	0.2	0.2	0.2	0.2
IND	6.5	5.8	6.9	5.6	4.9	4.6	6.5	5.8
IND/END	1.5	1.5	1.4	1.3	1.2	1.2	1.1	1.3
ETD	2.0	3.3	4.7	2.8	2.4	3.6	3.5	2.1
FeL	24.4	24.6	35.7	26.9	26.6	26.5	30.2	25.6
TiL	28.2	28.6	38.0	29.3	29.3	28.6	36.5	26.7
TaL	17.7	16.5	22.4	16.1	16.7	17.6	20.5	18.8
FL	23.8	22.8	32.2	23.9	24.4	22.4	30.3	23.3
HaL	19.1	19.1	25.5	17.3	17.2	16.7	21.2	17.6
FaL	15.1	14.1	16.9	11.2	11.3	10.0	16.0	15.6
UaL	10.0	11.5	14.8	14.9	14.8	11.7	13.9	13.1
FeL/SVL	0.5	0.5	0.5	0.5	0.5	0.5	0.4	0.4
FL/SVL	0.5	0.4	0.5	0.4	0.4	0.5	0.4	0.4
HaL/SVL	0.4	0.3	0.4	0.3	0.3	0.3	0.3	0.3
1FL	7.5	7.5	10.0	9.1	9.5	8.1	12.1	10.3
2FL	10.2	10.4	13.5	11.3	11.8	9.6	13.5	11.2
3FPW	3.2	2.9	4.2	2.9	3.1	2.9	4.2	3.7
3FW	1.2	1.7	1.8	1.3	1.3	1.6	2.3	1.6
3FL	12.8	13.9	19.8	12.9	13.4	12.9	14.0	12.0
4FPW	3.1	1.9	4.2	2.8	3.0	2.7	4.2	3.4
4FW	1.7	1.8	1.9	1.5	1.5	1.8	2.3	1.9
4FL	9.7	10.8	14.1	10.7	10.3	8.9	10.8	8.9
3FPW/3FW	2.7	1.7	2.4	2.2	2.3	1.9	1.8	2.3
4FPW/4FW	1.8	1.1	2.2	1.9	2.0	1.5	1.8	1.8
3FL/SVL	0.2	0.3	0.3	0.2	0.2	0.3	0.2	0.2
3FL/HaL	0.7	0.7	0.8	0.7	0.8	0.8	0.7	0.7
4FL/HaL	0.5	0.6	0.6	0.6	0.6	0.5	0.5	0.5
1TL	6.9	8.1	13.0	10.0	11.0	7.1	10.0	11.6
2TL	9.2	10.7	14.5	11.7	11.8	9.9	15.2	13.4
3TL	16.8	14.4	21.2	17.4	15.8	14.2	23.3	17.1
4TPW	2.8	2.5	3.7	2.5	2.4	1.7	3.6	3.1
4TW	1.3	1.4	2.6	1.4	1.5	1.0	2.2	2.1
4TL	20.1	18.0	26.5	22.2	17.3	19.6	26.4	22.8
5TL	14.5	14.6	19.0	17.9	15.0	13.6	19.3	18.2
4TL/FL	0.8	0.8	0.8	0.9	0.7	0.9	0.9	1.0

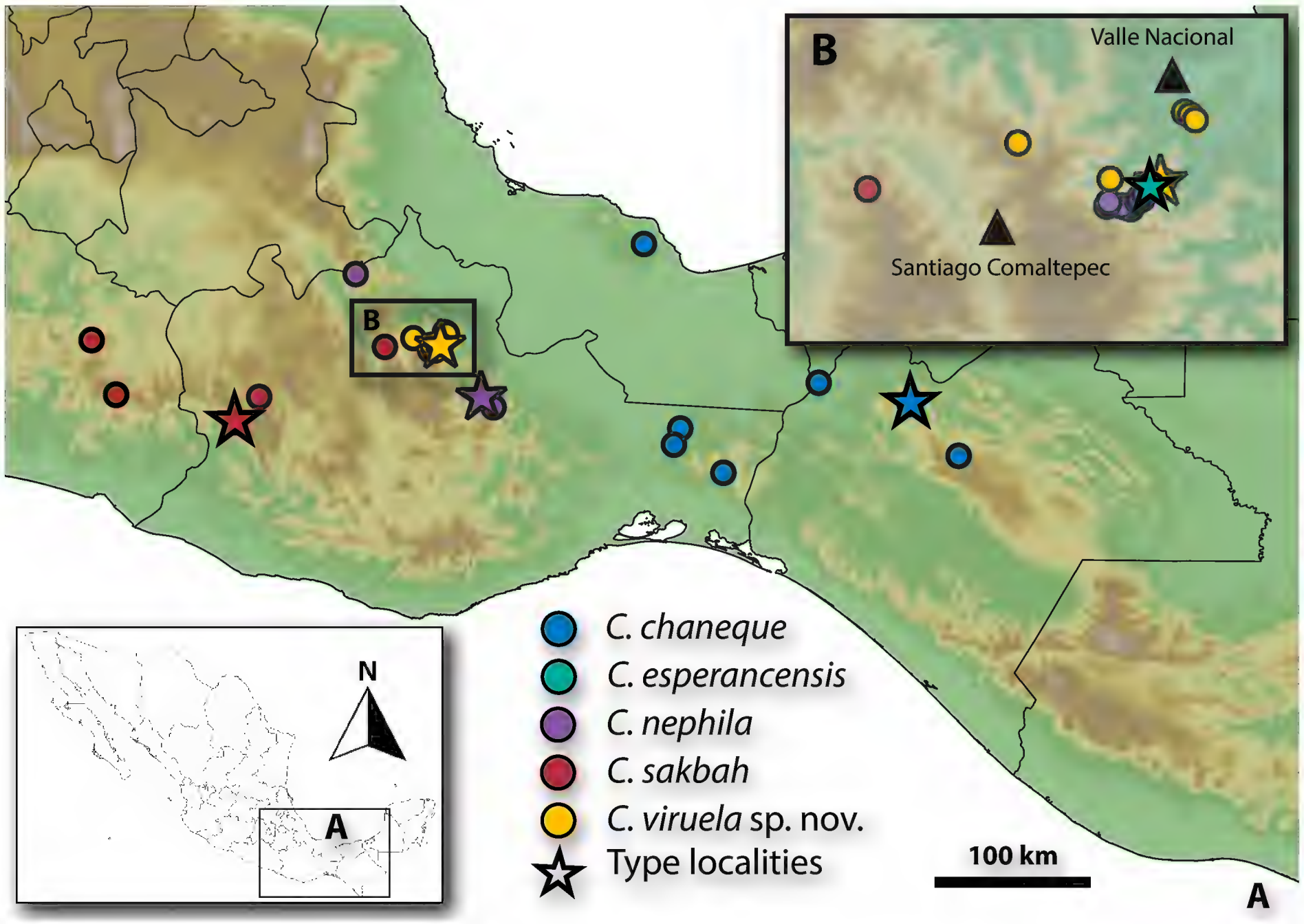

Figure 7. Distribution map of species of *Charadrahyla* in southeastern Mexico. Diamonds represent type localities. See materials and methods for source of records.

Figure 8. Photo of the locality and habitat at the type-locality of *Charadrahyla viruela* sp. nov. near La Esperanza, Municipio de Santiago Comaltepec, Oaxaca, Mexico.

stream vs. upstream), and thus reported different elevations and found different species. It should be noted that every specimen examined by us that was collected at "4.2 km S of Vista Hermosa" after 1990 is clearly identifiable as *C. viruela*. Of the 35 referenced specimens, we reviewed all 24 adult and juvenile specimens in photos; 11 are larva and we made no intent to identify those. Of the 24 specimens, three (KU 54335, 86966–67) are referrable to *C. viruela* based on the photographs, 17 are identifiable as *C. nephila* (see Appendix 1), and two are juveniles in bad states of preservation (UTA 3074–75) and impossible to assign to a species from photographs. One specimen (KU 71126) from "Arroyo Buena Vista" at 6.5 km S of Vista Hermosa, Oaxaca has intermediate characters between both species and may be a hybrid. This specimen possesses an untuberculate dorsum and the immaculate white spots on the chest which are characteristic of *C. nephila*, but it also presents the brown squarish blotches on the throat and the large lateral blotch pattern of *C. viruela*. These species are readily diagnosable in life, but unfortunately there are no photos of these specimens in life available.

Redefinition of *Charadrahyla nephila*

Charadrahyla nephila (Mendelson & Campbell, 1999)

Hyla chaneque—Duellman (1970): 440, in part.
Hyla chaneque—Adler and Dennis (1972): 1–19, in part.
Hyla chaneque—Caldwell (1974): 1–197, in part.
Hyla chaneque—Flores-Villela (1993): 1–73, in part.
Hyla chaneque—Toal and Mendelson (1995): 1–20, in part.
Hyla nephila—Mendelson and Campbell (1999): 80, in part.
Hyla nephila—Duellman (2001): 937–939, in part.
Charadrahyla nephila—Faivovich, Haddad, García, Frost, Campbell, Wheeler (2005): 100.
Charadrahyla nephila—Stuart, Hoffmann, Chanson, Cos, Berridge, Ramani, Young (2008): 241.
Charadrahyla nephila—Campbell, Blancas Hernández, Smith (2009) 287–295, in part.
Charadrahyla nephila—Heimes (2010): 80–88.
Charadrahyla nephila—Delia, Whitney, Burkhardt (2013): 1405–1414, in part.
Charadrahyla nephila—Caviedes-Solis, Vázquez-Vega, Solano-Zavaleta, Pérez-Ramos, Rovito, Devitt, Heimes, Flores-Villela, Campbell, Montes de Oca (2014): 229–241, in part.
Charadrahyla nephila—Lamoreux, McKnight, Cabrera-Hernández (2015): 105–160, in part.
Charadrahyla nephila—Canseco-Márquez, Ramírez-González, González-Bernal (2017): 64–72, in part.
Charadrahyla nephila—Jiménez-Arcos, Calzada-Arciniega, Alfaro-Juantorena, Vázquez-Reyes, Blair, Parra-Olea (2019): 371–385, in part.
Charadrahyla nephila—León-Gómez (2019): 1–79.

Holotype. *Hyla nephila* Mendelson & Campbell, 1999: 80. Type locality "5.8 km W Totontepec, Sierra Mixe, Oaxaca, Mexico, 2,103 m asl 17°13'N, 96°03'W". UTA A-5769, adult male, collected by Jonathan A. Campbell, on 04 April 1978.

Diagnosis. A moderately sized *Charadrahyla* (males 53.7–61.9 mm, females to at least 79.3 mm) with a relatively short snout, rounded in dorsal and lateral profile; smooth dorsal skin and a brown dorsal ground coloration, with large dark brown dorsal blotches, which are varyingly sprinkled with green iridophores, on some specimens green coloration is very sparse, while on others green iridophores combine to form large areas of green coloration on the large brown dorsal blotches; dorsal tubercles absent, thus giving dorsal ground coloration a clean look; the overall combination of the dorsal patterning gives the frog a "military camouflage" appearance. It can be distinguished from all other species of *Charadrahyla* by the combination of external tympanum visible; a lack of a rostral keel; lack of axillary membrane; lack of conical tubercles on either side of cloacal opening; lack of hypertrophied webbing between first and second toe; and by the absence of tuberculate dorsal skin and the distinctively plain copper-colored iris lacking dark reticulations. Other identifying characteristics include distinctly rounded subarticular tubercles on all fingers; upper lip same color as lateral portions of body, no distinctive pale blotch present on lip posterior to eye; ventral coloration gray or brown colored, not while or cream, with 1–7 immaculate white spots present on chest and abdomen.

Comparisons. *Charadrahyla nephila* can be distinguished from all currently recognized species of *Charadrahyla* by the combination of the following characteristics: external tympanum present; dorsal skin smooth, no tubercles; snout short in both adult males and females; snout short and rounded in both dorsal and lateral profiles; no rostral keel; subarticular tubercles on fingers rounded, not bifid; no axillary membrane; no hypertrophied webbing between first and second toes; no conical cloacal tubercles; possessing nuptial excrescenses in adult males; unique dark green blotches on a tan or pale brown dorsal coloration with dark green dorsal blotches, which gives the frog an overall "military camouflage appearance"; lack of well-defined and highly contrasting dorsolateral blotches, dorsal lateral pattern and coloration similar to other blotches on dorsum; iris copper, clean and un-patterned, without black reticulations. Phenotypically this species appears to be most similar to *C. viruela* and *C. chaneque*, the former occurring sympatrically or nearly sympatrically at moderate elevations of the Sierra Juárez of northern Oaxaca. *Charadrahyla nephila* differs from *C. viruela* (Comparisons in Figs 4, 5) by possessing smooth dorsal skin on the entire dorsum (vs. tuberculate skin); longer snout, which is rounded in lateral profile (vs. shorter snout, which is truncate in lateral profile); nostrils not flared (vs. flared nostrils); subarticular tubercles rounded, not bifid (vs. subarticular tubercles on some fingers distinctly bifid); finger discs same size in both sexes (vs. sexual dimorphism in finger-disc size); dorsal ground coloration brown, with large dark brown dorsal blotches, which are varyingly sprinkled with green iridophores, on some specimens green coloration is very sparse, while on others green iridophores combine to form large areas of green coloration on the large brown dorsal blotches; dorsal tubercles absent, thus giving dorsal ground coloration a clean look; the overall combination of the dor-

sal patterning gives the frog a "military camouflage" appearance (vs. dorsal ground coloration emerald green, with large dark green dorsal blotches, which paler green interiors and darker green outlines, and dark green spots on the pale interior; numerous dorsal tubercles are reddish-brown or tan; some tan blotching is present dorso-laterally; the overall combination of the dorsal patterning gives the frog a "mossy" appearance); no distinctive white blotch on upper lip (vs. distinctive white or whitish-yellow blotch on upper lip posterior to eye); iris solid copper coloration (vs. iris golden, with heavy black reticulations, giving it a starry appearance); subarticular tubercles rounded (vs. distinctly bifid on some fingers); distinctive white blotch on upper lip (vs. no distinctive white blotch on lip); throat, chest, and abdomen is brown, with 1–7 immaculate white spots present on throat or chest and no large tan blotches on throat; and no highly contrasting lateral blotches present (vs. distinct, regular, well defined and highly contrasting lateral blotches compared to other blotches on dorsum).

Charadrahyla nephila differs from *C. chaneque* as follows: smooth dorsal skin (vs. dorsal skin smooth or slightly tuberculate); dorsal ground coloration brown, with large dark brown dorsal blotches, which are varyingly sprinkled with green iridophores, on some specimens green coloration is very sparse, while on others green iridophores combine to form large areas of green coloration on the large brown dorsal blotches; dorsal tubercles absent, thus giving dorsal ground coloration a clean look; the overall combination of the dorsal patterning gives the frog a "military camouflage" appearance (vs. dorsal ground coloration tan to pale brown, with larger dark brown dorsal blotches, which for the most part lack green iridophores; green iridophores widespread on dorsal ground coloration, giving some specimens a greenish appearance; dorsal tubercles present, same color as skin around them). Comparisons between *C. nephila*, *C. viruela*, and *C. chaneque* are given in Table 2.

Charadrahyla nephila can be distinguished from *C. esperancensis* by its lack of a rostral keel (vs. distinct rostral keel present), much shorter snout, rounded from a lateral profile (vs. longer snout, protruding in lateral profile), presence of large blotching on the lateral flanks and lack of yellow reticulation (vs. distinctive brown and yellow reticulations, no blotches); unique brown dorsal coloration with large dark brown and green dorsal blotches (vs. dorsal coloration solid brown coloration with some darker brown spots).

The widespread *Charadrahyla taeniopus* is a highly variable species, and some of its pattern classes may be readily confused with *C. nephila*. However, *C. nephila* can be distinguished from *C. taeniopus* by smaller adult size, with adult males 53.3–61.9 mm (vs. up to 65.9); short, rounded snout in both adult male and female lateral profile (vs. rounded snout in lateral profile in females and subadults, elongated and acuminate in adult males); and lacking enlarged testes (vs. enlarged).

Charadrahyla nephila can be distinguished from *C. altipotens* by its unique brown dorsal coloration with large dark brown and green dorsal blotches, (vs. dorsal coloration brown or green, usually green, with no large dorsal blotches present); snout rounded in lateral profile (vs. accuminate) and axillary membrane absent (vs. present); nuptial excrescences present in adult males (vs. absent) and testes not enlarged (vs. enlarged).

Charadrahyla nephila can be distinguished from *C. sakbah* by its dorsal coloration with dark brown and green spots, giving a high-contrast dorsal patterning with an overall "military camouflage look" (vs. dorsal coloration brown, with brown or green blotching, pattern not high contrast); lack of axillary membrane (vs. present); lack of hypertrophied webbing on the foot of males (vs. present); lack of conical tubercles around vent (vs. present); lack of enlarged testes (vs. enlarged) and a dark ventral coloration throat, chest and abdomen is brown, with 1–7 immaculate white spots present on throat or chest (vs. white or pale cream abdomen, with no immaculate white spots).

Charadrahyla nephila can be distinguished from the two larger species in Guerrero, *C. trux* and *C. tecuani*, by lacking hypertrophied webbing on the foot of adult males (vs. present); rounded snout from a lateral profile (vs. acuminate or slightly acuminate); a dark ventral coloration throat, chest and abdomen is brown, with 1–7 immaculate white spots present on throat or chest (vs. yellow creamy brown abdomen, with no immaculate white spots).

Finally, *C. nephila* differs from the two smaller members of the genus (*C. pinorum* and *C. juanitae*) by presenting an external tympanum (vs. lacking external tympanum); its larger size of 53.7–61.9 mm in males (vs. 27.7–35.8 mm); subarticular tubercles on fingers rounded, not bifid (vs. subarticular tubercles on fingers bifid); lack of axillary membrane (vs. presence of axillary membrane, albeit small); unique brown dorsal coloration with large dark brown and green dorsal blotches (vs. dorsal coloration pale brown with darker brown blotches, no green and no military "camouflage" appearance); cloacal tubercles indistinct (distinctive cloacal tubercles present both above and below cloacal opening).

Distribution and ecology. *Charadrahyla nephila*, as now understood, is endemic to Oaxaca, where it occurs at moderate to high elevations of the Sierra Mixe, Sierra Juárez, and Sierra Mazateca in the Sierra Madre Oriental region in the northern part of the state. It is predominately limited to windward slopes of the Atlantic drainage but may occur further inland as well where proper conditions exist such as in the Sierra Mazateca and adjoining portions of highlands in the states of Puebla and Veracruz. In the Sierra Mixe it is known from several localities at elevations from 1,660–2,175 m asl in the vicinity of Totontepec Villa de Morelos and Santiago Zacatepec. In the Sierra Juárez it occurs along Hwy. 175 from as low as "1 km S of La Esperanza" (1,665 m) up to "16.6 km S o f Vista Hermosa" (2,150 m). It has been collected in the municipalities of Santiago Comaltepec and Ixtlán de Juárez. Finally, in the Sierra Mazateca it has only been collected along Hwy. 182 at elevations between 2,150–2,180 m asl in the Municipio de Santa María Teopoxco. All localities are along steep cascading streams in montane upper montane cloud forest or very moist pine-oak woodland.

Referred specimens: See Appendix 1.

The tadpoles and juveniles of *Charadrahyla viruela* and *Charadrahyla nephila*

The following description is based on a single tadpole of *C. viruela* (CNAR 37136) collected on 21 April 2018 in GS 35 from a stream about 500 m (airline) NE of La Esperanza, Municipio de Santiago Comaltepec, Oaxaca (Fig. 9A; mouthparts in Fig. 10). Measurements (mm): total length 63.1; body length 20.1; tail length 423.0; tail muscle height 8.6; maximum tail height 11.8; tail muscle width 6.2; interorbital distance 8.3; internarial distance 7.2; oral disc diameter 6.6.

Body ovoid in dorsal view; in lateral view higher posteriorly than anteriorly; snout rounded in dorsal and lateral view; eyes moderate in size, positioned and directed dorsolaterally; interorbital distance slightly larger than internarial distance; nostrils small, oval, directed dorsolaterally. Spiracle single, sinistral, short, opening near midbody. Vent tube dextral. Tail long, 2.1 times the body length. Caudal musculature robust, highest at junction with body, rounded at tip. Dorsal fin originating on the trunk of the body, at distal fifth of trunk, and becomes taller at mid-length of tail. Ventral fin about same size as the dorsal fin, and similar in shape. Neuromasts in the lateral line system visible. Oral disc large, ventral in position, not emarginate, bordered by two complete rows of marginal papilla; numerous submarginal papillae present laterally. Labial keratodont row formula 2(2)/4(1), first anterior row slightly longer than second, second row with a medial gap; first three lower keratodont equal in size, longer than the fourth lower row, first row with very narrow medial gap, about width of approximately seven keratodonts. Upper jaw moderately wide, lateral processes taper abruptly posterolaterally; lower jaw shallowly V-shaped; both jaws with large serrations.

In life, body pale brown dorsally with pale green spots; ventrum brown, darker than dorsum; caudal musculature yellowish, with a few cream spots in the first third of the caudal musculature; caudal fins cream with few spots on dorsal fin. Iris golden, with black reticulations. Other tadpoles of *C. viruela* were observed in the same stream with the same coloration pattern (Fig. 9B, GS 29). Tadpoles in GS 41, (Fig. 9C) have dark spots and gold markings on the caudal musculature and dorsal and ventral fins; the body also has dark spots, and green tones beginning to appear on the dorsal part of head, and canthal bar is visible; hind limbs well developed and barred, and tubercles on legs beginning to become noticeable. A GS 42+ metamorph (Fig. 9D) was observed at night perched on a branch about 50 cm from the ground on the edge of the stream. At this stage the adult characteristics of the species start to become evident, including dark spots on the body against an emerald green ground coloration, and the black and gold reticulations in the eye, which cause the "starry-eyed" appearance. The upper lip displays the white markings characteristic of adult specimens. The tuberculate dorsal texture is beginning to become evident, being more pronounced on the forelimbs and hindlimbs.

The description of the tadpole of *Charadrahyla nephila* is given in Duellman (1970, 2001) and Mendelson and Campbell (1999). In life, tadpoles of *C. nephila* have a dark brown body dorsally, yellowish tan laterally; dark gray ventrally; body with pale green streaks; caudal fins transparent, with scattered dark brown spots; xanthophores

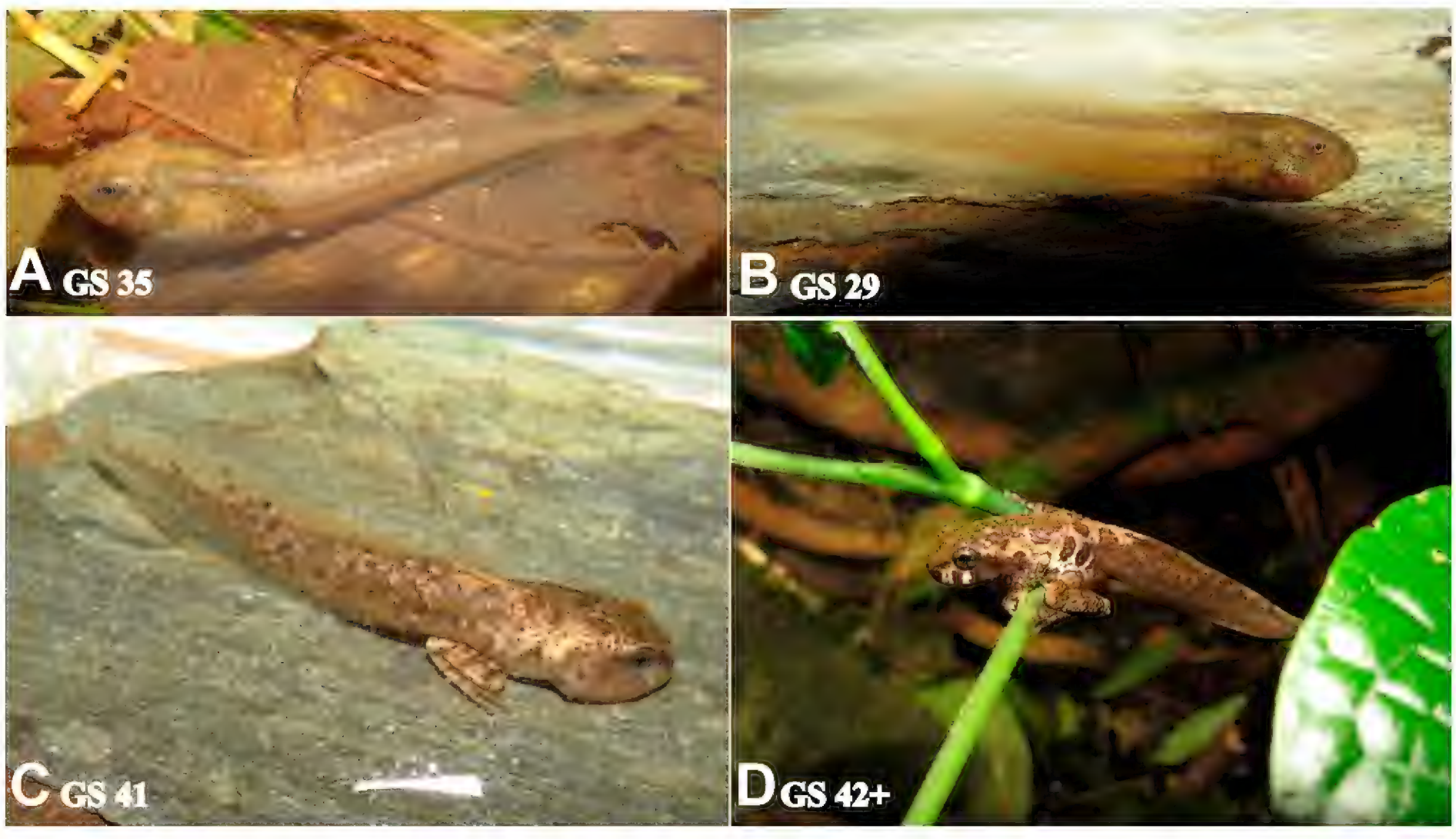

Figure 9. Photos of tadpoles and recent metamorphs of *Charadrahyla viruela* sp. nov. from a small stream located at 0.5 km (airline) NE of La Esperanza, Municipio de Santiago Comaltepec, Oaxaca. The GS numbers are in reference to developmental stages as defined by Gosner (1960).

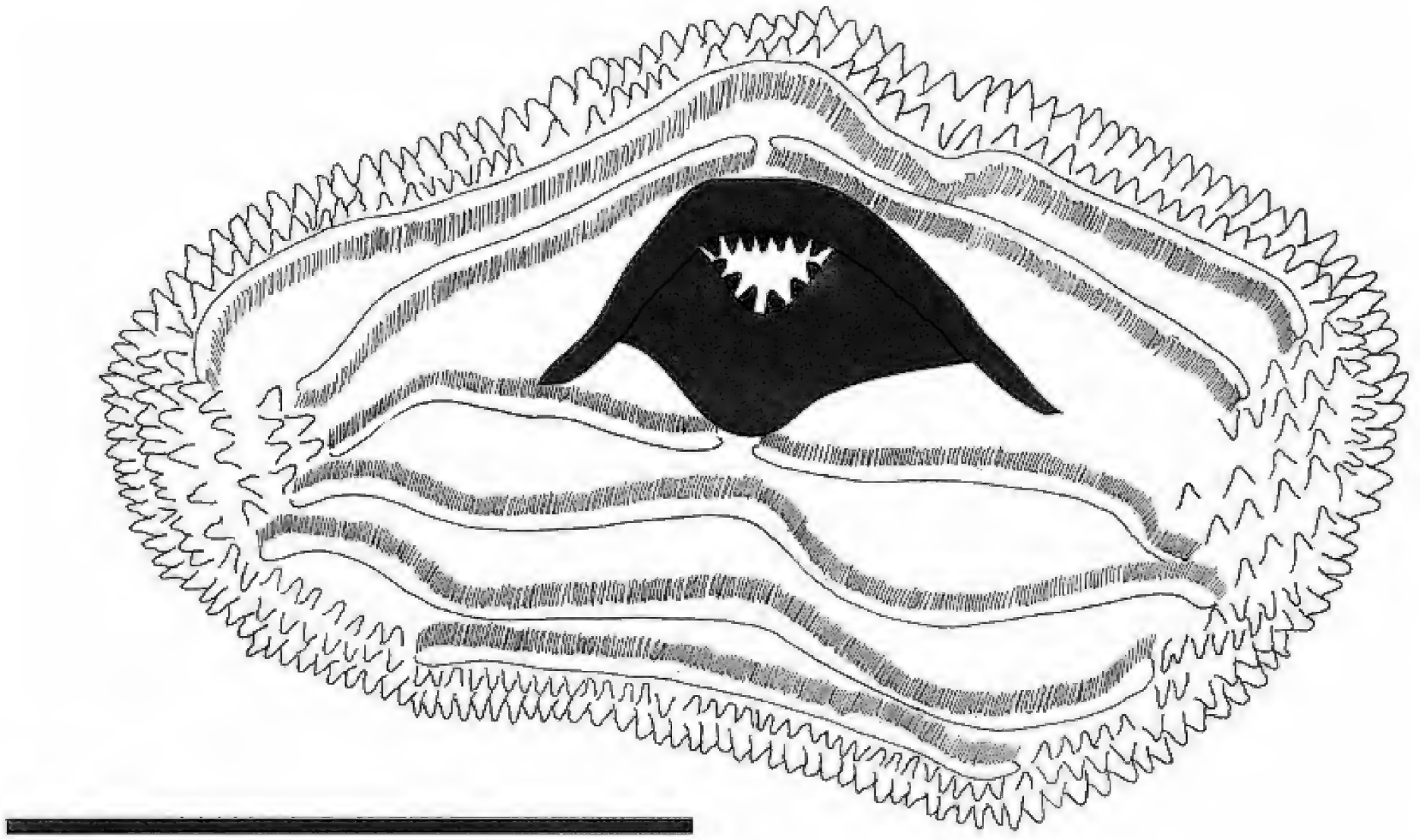

Figure 10. An illustration of the mouthparts of a tadpole (CNAR 37136) of *Charadrahyla viruela* sp. nov. from 0.5 km (airline) NE of La Esperanza, Municipio de Santiago Comaltepec, Oaxaca. Illustration by Luis Canseco-Márquez. Black line represents 4.0 mm.

forming distinct yellow spots on caudal musculature and fins, forming yellow edge on dorsal fin; iris yellow. In preservative, entire body brown; caudal musculature yellowish-tan; caudal fins transparent with scattered small, brown spots (Duellman, 1970; Mendelson and Campbell 1999).

We found several tadpoles of *C. nephila* in a stream approximately 3 km (airline) SW of La Esperanza, Municipio de Santiago Comaltepec, Oaxaca. Tadpoles from this stream were kept in captivity and are shown in Fig. 11. The morphology of the oral disc is similar to that of the *C. viruela* tadpoles (Fig. 11I), but the color pattern (Fig. 11B, C, E) is as mentioned by Duellman (1970) for *C. nephila* (see above), except one tadpole with presence of dark oval spots on the body (Fig. 11A). Two metamorphic juveniles were observed in the field perched about 1 m above the ground next to the stream at GS 44 and 45 (Fig. 11J, M, respectively). Dorsal ground coloration in those two juveniles is lemon green, showing some irregular black spots on the back and markings on the sides; the limbs are already barred, and the pupil of the eye is yellow (Fig. 11J, M). A tadpole at GS 38 (Fig. 11C) already had well-developed toes. By GS 42 (Fig. 11D), all four limbs are already barred and fully developed, the tail beginning to reabsorb, and the background color of the body beginning to turn green. Another sequence from tadpole to juvenile is observed in Fig. 11E–H; at GS 41 (Fig. 11E), the tadpole has a total length of 60.5 mm, barred and well-developed hind limbs, the forelimbs are already visible, although they are still inside the body; upon reaching GS 44 (Fig. 11F, G) all the limbs have developed, and dark spots are present on the body on a green background. At reaching GS 45 (Fig. 11H), the tail has been almost completely reabsorbed, and at GS 44 (Fig. 11J), it attains a length of 19.7 mm. Upon reaching GS 46 (Fig. 11K), the metamorphosis is complete, and the juvenile has SVL = 29.0 mm. During the process of completing the metamorphosis (Fig. 11M), the juveniles have changes in color pattern. As they grow, they maintain green coloration on the sides of the body (Fig. 11N) and until reaching approximately 40 mm in length they can have a pale brown background body color with dark spots all over the body (Fig. 11O). Some juveniles, also at GS 46, may show green coloration on the sides of the body, including the side of the head, and brown on the back (Fig. 11L). The juveniles of *C. nephila* have a noticeably unpatterned iris with no black reticulation, no "starry-eyed" appearance, and are readily distinguished from juveniles of *C. viruela* by the pattern and coloration of their iris.

Charadrahyla sakbah in the Sierra Juárez, Oaxaca

Charadrahyla sakbah is known from the Sierra Madre del Sur of Guerrero and Oaxaca, including two localities in eastern Guerrero (Bautista del Moral et al. 2021) and three localities in western and central Oaxaca (Jiménez-Arcos et al. 2019, Mata-Silva et al. 2019, Palacios-Aguilar et al. 2022). Here, we report on a population from across the dry tropical lowlands of the Valle de Oaxaca in the interior drainage of the Sierra Monteflor section of the Sierra Juárez (Sierra Madre Oriental), which was formerly mentioned in Canseco-Márquez & Gutiérrez-Mayén (2010) as "*Charadrahyla* sp."

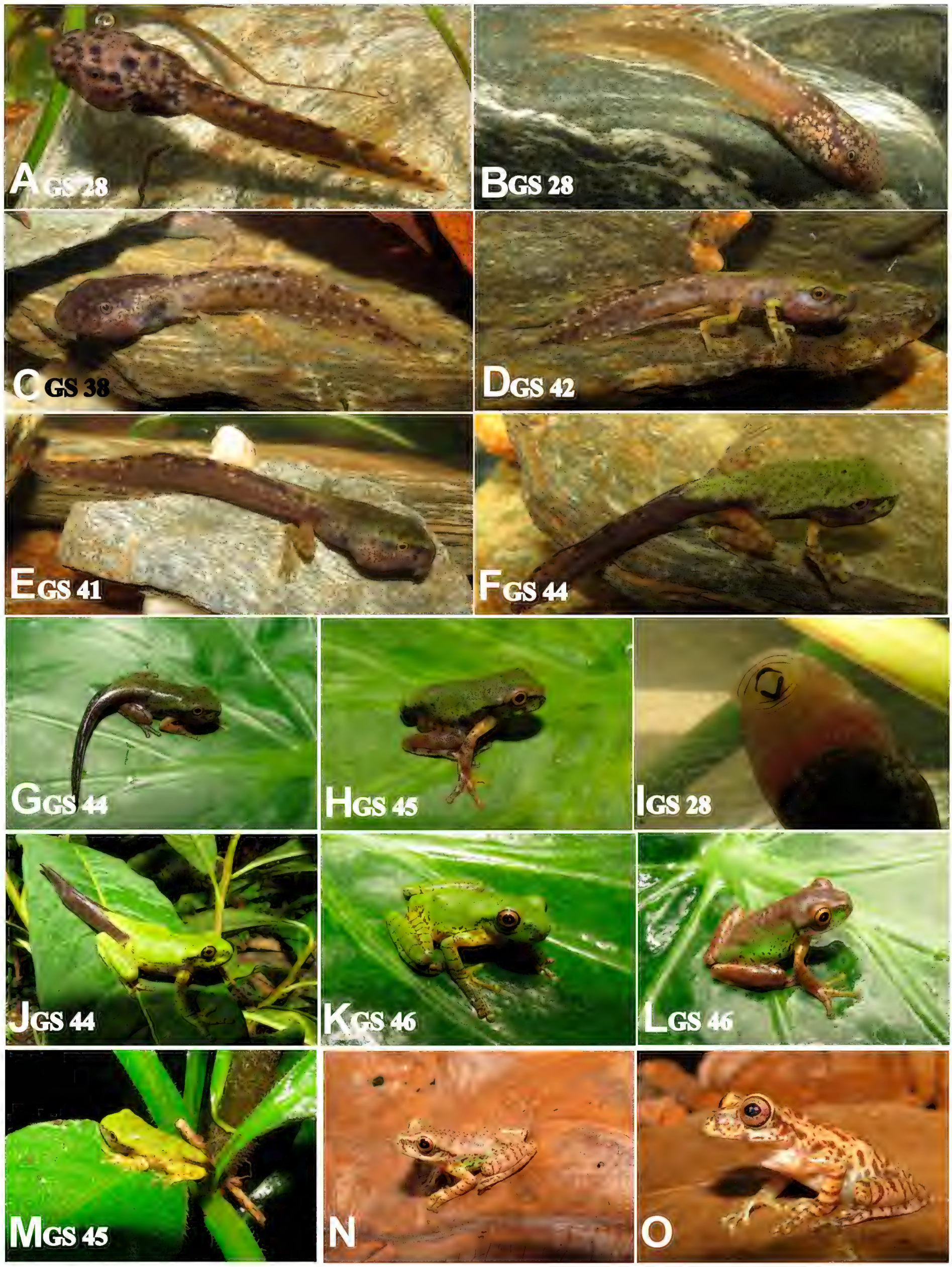

Figure 11. Photographs of tadpoles and recent metamorphs of *Charadrahyla nephila* from a stream approximately 3 km (airline) SW of La Esperanza (approx. 9.1 km S of Vista Hermosa), near the San Bernardo Trail, Municipio de Santiago Comaltepec, Oaxaca. The GS numbers are in reference to developmental stages as defined by Gosner (1960).

Referred specimens: Adult male (MZFC 21980; Fig. 12B), collected in stream on Cerro Monteverde, approximately 12.6 km (airline) NE of Zoquiápam Boca de los Ríos, Municipio de San Juan Bautista Atatlahuca (17.6170, -96.7519°; 2,476 m asl; datum = WGS84), Oaxaca, Mexico, on 09 March 2008 by Luis Canseco-Márquez; adult female (MZFC 21979; Fig. 12A), subadult male (MZFC 21978) and two juveniles (MZFC 21981–82 Fig. 12C), collected in stream on Cerro Monteverde, approximately 11.7 km (airline) NE of Zoquiápam Boca de los Ríos, Municipio de San Juan Bautista Atatlahuca (17.6183, -96.7576°; 2,233 m. asl; datum = WGS84), Oaxaca, Mexico, on 08 March 2008 by Luis Canseco-Márquez.

This record extends the known range of *Charadrahyla sakbah* (Fig. 7) approximately 104 km to the northeast from the nearest reported locality in the vicinity of Tlaxiaco, Oaxaca (Palacios-Aguilar et al. 2022). This is a first record for the Municipio de San Juan Bautista Atatlahuca, as well as the first record for the Sierra Monteflor and the entire Sierra Juárez complex. The Sierra Monteflor is located in the highlands of northern Oaxaca and comprises the westernmost portion of the Sierra Juárez complex, which is part of the Sierra Madre Oriental. These specimens were located in two streams in pine-oak forest and humid oak woodland on Cerro Monteverde. During the month of March, on one night, 30 individuals were observed on branches along these streams. A later visit to the area during the rainy season (August) did not produce any individuals in the same streams where they were observed in March. Other anurans that were observed in the same streams with *C. sakbah* include *Sarcohyla cyclada* and *Craugastor pygmaeus*.

We shared tissue of these populations with Itzue Caviedes-Solis, to include them in a molecular phylogeny of Middle American hylids. We are unable to share these results herein, but they were confirmed to represent *C. sakbah* (I. Caviedes-Solis, pers. comm.). Measurements for specimens of *C. sakbah* from the Sierra Monteflor are included in Table 4.

Discussion

Availability of genetic material

Several recent studies have questioned the use of mitochondrial DNA alone for species delimitation in the absence of nuclear data (Reyes-Velasco 2024; Wüster 2025). However, this view is not universal, particularly in amphibians and within Hylidae, where numerous studies have successfully used mitochondrial loci to delimit species and infer evolutionary relationships (Stöck 2008; Wiens et al. 2010; Yan et al. 2020; Warwick et al. 2021). Moreover, the utility of any genetic marker depends largely on the availability of comparative material. In groups composed of rare and difficult-to-collect species, such as *Charadrahyla*, comprehensive sampling across loci is often impractical. Currently, little nuclear sequence data is available for *Charadrahyla* and *Megastomatohyla*. However, there are several sequences available in Genbank, and also some published by Caviedes-Solis et al. (2020), which we included in this study. In contrast, mitochondrial loci 12S and 16S represent the most widely sampled genes for amphibians and provide the only datasets with sufficient comparative coverage to allow meaningful phylogenetic inference and estimation of interspecific genetic distances within the genus.

Charadrahyla nephila

Mendelson and Campbell (1999) described *Charadrahyla nephila* from the Sierra Mixe, Sierra Juárez, and Sierra de Zongolica (Sierra Mazateca) of Oaxaca. They assigned 52 specimens as paratypes, with six originating from the Sierra Mixe, two from the Sierra de Zongolica, and the rest from a corridor along Highway 175 in the Sierra Juárez. Apparently, they failed to detect the differences between *C. viruela* and *C. nephila* in their revision of preserved specimens, and our review of photographs of many of their type specimens suggests that they had included both *C. nephila* and *C. viruela* among their paratypes. However, it is noteworthy that the type-locality they designated is in the Sierra Mixe, where to date only *C. nephila* has been collected. We reviewed detailed photographs of the type-specimen, and it is clear to which form the name *C. nephila* applies. While we assigned four specimens of the type series of *C. nephila* to *C. viruela*, it is noteworthy that we only restrict the distribution of *C. nephila* elevationally, and specimens assignable to *C. nephila* remain from the Sierra Mixe, Sierra Juárez, and Sierra Zongolica.

Table 4. Morphometric variation of *Charadrahyla sakbah* in the Sierra Monteflor, Sierra Juárez, Oaxaca.

	MZFC 21980 (Male)	MZFC 21978 (Subadult Male)	MZFC 21979 (Female)	MZFC 21981 (Juvenile)	MZFC 21982 (Juvenile)
SVL	75.3	60.9	80.8	41.7	38.7
HL	21.2	18.3	21.9	12.2	11.7
HW	22.8	19.7	25.6	13.7	12.70
ED	7.4	6.9	8.20	4.6	4.4
TW	2.25	2.8	3.8	1.6	1.4
TL	40.6	32.7	42.6	21.8	19.5
TotFL	35.4	26.6	38	17.4	14.9

Figure 12. Variation in color and pattern of *Charadrahyla sakbah* from the Sierra Monteflor, Oaxaca. **A.** Adult female (MZFC 21979); **B.** Adult male (MZFC 21980); **C.** Adult male (MZFC 21978); **D, E.** Subadult males (not collected); **F, G.** Adults of unknown sex (not collected). All from the same stream at approximately 11.7–12.6 km (airline) NE of Zoquiápam Boca de los Ríos, Municipio de San Juan Bautista Atatlahuca, Oaxaca, Mexico.

Diversity and ecology of *Charadrahyla* in the Sierra Juárez

With the description of *C. viruela*, a total of four species of *Charadrahyla* are now known from the Sierra Juárez in northern Oaxaca. Two closely related species, *C. sakbah* and *C. esperancensis* occur on either side of the high continental divide in the Sierra Juárez, with *C. sakbah* inhabiting the relatively drier interior portions of this complex (Sierra Monteflor, see below) and *C. esperancensis* occurring at medium elevations on the humid windward slopes (La Esperanza). *Charadrahyla viruela*, *C. nephila*, and *C. esperancensis* all occur on the humid windward northern slopes of the Sierra Juárez. *Charadrahyla viruela* and *C. esperancensis* apparently occur syntopically in the same stream at 4.2 km S of Vista Hermosa, Oaxaca. The closely related *C. viruela* and *C. nephila* both occur on the humid windward slopes of the Sierra Juárez, but they seem to be, for the most part, separated from each other by elevation, with *C. viruela* occurring at lower elevations (600–1,725 m asl) and *C. nephila* occurring at higher elevations (1,600–2,250). However, our revision of museum records indicates that both species have been collected from the same reported locality, at least at one locality (referenced as 4.2 km S of Vista Hermosa, Municipio de Santiago Comaltepec, Oaxaca). However, we have visited this locality on many occasions and have only collected *C. viruela*. Older localities tend to be more vaguely described, and many localities are given generally with the road distance given of where a stream crosses the road, not necessarily where the specimen was collected. The extreme topography of Oaxaca, coupled with heavy rainfall, can cause very different habitats to occur within close proximity of each other on the same slope. Similar species can be isolated by habitat types, within a few hundred meters of each other. Similar narrow elevational separation at the same locality on steep slopes in Oaxaca has been previously observed in the Sierra Madre del Sur with the separation of *Ophryacus sphenophrys* (cloudforest) and *Ophryacus undulatus* (oak forest) (Grünwald et al. 2018). To the best of our knowledge, we ignore if other cases of isolation along an elevation gradient have been reported for other Mexican amphibians.

Individuals of *C. viruela* have been found active throughout the year at night along streams, perched on branches ranging from 30 cm to 3 meters above the ground, and several individuals have been observed on branches within the splash zones of waterfalls. The percentage of humidity in the environment at the stream located at 4.2 km S of Vista Hermosa (known locally as Arroyo Tres Manantiales) typically remains between 70–77% and the locality maintains an ambient temperature of 18–19 °C (LCM pers. data).

Approximately 50 individuals of *C. viruela* were recorded over the course of a year during 2018 and 2019 in a stream near Santa Cruz Tepetotutla, Municipio de San Felipe Usila (R. Simón-Salvador pers. data), and were recorded at streams with limited human activity, suggesting that they may be sensitive to human presence (Simón-Salvador et al. 2021). León-Gómez (2019) in his study about the effect of cloudforest disturbance on four native species at La Esperanza, observed that habitat characteristics for *C. viruela* include streams where rocks on the stream margins and stream bottom dominate the channel cover, high densities of trees, and streamside shrubs at least two meters tall with branches approximately 1.4 cm in diameter to serve as perches.

Other anuran species that we observed in the streams with *C. viruela* near La Esperanza include: *Charadrahyla esperancensis*, *Duellmanohyla ignicolor* (Duellman 1961), *Exerodonta abdivita* (Campbell & Duellman, 2000), *Ptychohyla zophodes* Campbell & Duellman, 2000, *Craugastor polymniae* (Campbell, Lamar, and Hillis 1989), and *C. spatulatus* (Smith 1939). Near Santa Cruz Tepetotutla other species of anurans observed in the same stream as *C. viruela* include *Duellmanohyla ignicolor*, *Exerodonta abdivita*, *Quilticohyla acrochorda* (Campbell & Duellman, 2000), *Triprion spinosus* (Steindachner 1864), *Hyalinobatrachium viridissimum* (Taylor 1942), and *Craugastor polymniae* (Simón-Salvador pers. comm.; Arreortúa et al. 2021).

Conservation priorities

Charadrahyla chaneque and "*Charadrahyla duellmani*"

Charadrahyla chaneque was described by Duellman (1961) from a stream at 6.2 km S Rayón Mescapala, in the highlands of northern Chiapas. Shortly thereafter, Lynch and Smith (1966) described *Charadrahyla duellmani* from two specimens collected at an unprecise locality in the Chimalapas north of Zanatepec, Oaxaca. Duellman 1970 synonymized *C. duellmani* with his concept of *C. chaneque*, which included both *C. nephila* and *C. viruela* at the time. Duellman (2001) revisited the known specimens of *C. chaneque*, after separating specimens of *C. nephila* (including several *C. viruela*) from his samples. At that time, *C. chaneque* was only known to him from seven specimens collected within 1 km radius of the type-locality, and the two mysterious specimens from the Chimalapas (KU 106296; UIMNH 56821). However, two more specimens (MZFC 18911–12) of *C. chaneque* were collected by LCM in May of 1995 at 330 m asl in Chalchijapa, Santa Maria Chimalapa, on the Atlantic versant of Oaxaca, and another recent record of *C. chaneque* (Ríos-Rodas et al. 2019) based on a metamorph specimen collected in Municipio de Huimanguillo, extreme southern Tabasco, whose identity was confirmed by genetic barcoding (L. Canseco-Márquez unpublished). The species was recently reported from the vicinity of Santa María Chimalapa, at a locality at ~260 m, which also represents the lowest elevation known for the species or for any member of the genus (García-Padilla et al. 2024). Furthermore, we are aware of one specimen (CNAR 18933) from Veracruz,

collected in the Sierra Santa Martha, Municipio de Soteapan, in the Los Tuxtlas region of that state. This specimen has been referred to as both *C. chaneque* and *C. nephila* (Pérez-Higareda 1981; López-Luna 2017), however we have examined this specimen and feel confident that it is assignable to *C. chaneque*.

The IUCN classifies *C. chaneque* as Vulnerable under criteria B1ab(iii) (assessed 2019), based primarily on a larger assumed range which stems from the past confusion about which specimens were *C. chaneque*. The species has been observed recently at its type locality in 2017 and 2024, and reported to occur in southern Tabasco in 2018 and near Santa Maria Chimalapa in 2021 (García-Padilla et al. 2024). The Chimalapas records from the Pacific versant are of questionable identity, and over 62 years old. The Atlantic versant records are likely *C. chaneque*, but are also over 30 years old. The Veracruz record is based on one specimen which was collected over 46 years ago, and has not been collected again despite recent efforts (CIG, LCM pers. data).

Three of the localities where *C. chaneque* has been collected recently are under extreme threat of habitat degradation due to small-scale and medium-scale agriculture, as well as siltation from slash and burn practices. We propose that *C. chaneque* be re-categorized as Endangered based on criterion B1ab(iii) as it can only be confirmed from four localities in the last 25 years and we estimate continuing decline observed, inferred, or projected in the area, and decline in extent and/or quality of habitat.

Charadrahyla nephila

Charadrahyla nephila was described by Mendelson and Campbell (1999) as occurring in the Sierra Mixe, Sierra Juárez, and the Sierra Mazateca in northern and central Oaxaca. Even with the description of *C. viruela* herein, and the assignation of several specimens formerly assigned to *C. nephila* to the new species, the distribution of *C. nephila* is still understood to encompass all three major mountain ranges. This species has not been reported from the Sierra Mazateca since before its description, but recent collecting efforts have produced one adult specimen (CIG pers. data).

The IUCN currently lists *C. nephila* as Endangered under criteria B1ab(iii) (assessed 2019). As we have no data that greatly modifies our understanding of the overall distribution or habitats for *C. nephila* we do not recommend an adjustment for this species. However, we do note that several species of *Charadrahyla* with much smaller ranges and likely more pressing threats to their existence (*C. esperancensis*, *C. chaneque*) are currently not awarded the Endangered category.

Charadrahyla sakbah

Charadrahyla sakbah was described by Jiménez-Arcos et al. (2019) from several specimens originating from one locality, in the northwestern portion of the Sierra Madre del Sur of Oaxaca. Its range was then extended within Oaxaca by Mata-Silva et al. (2019) and to eastern Guerrero by Bautista del Moral et al. (2021). Herein we have extended the range significantly into the Sierra Monteflor region of the Sierra Juárez in northern Oaxaca.

The IUCN currently lists *C. sakbah* as Endangered under criteria B1ab(iii) (assessed 2019). As the range of this species has been extended considerably since its last evaluation, we recommend a re-evaluation of its status with the new localities taken into consideration.

Charadrahyla viruela

The species described herein has a very limited range, known from 6 localities within a 23 km radius of the type-locality on the northern slopes of the Sierra Juárez, Oaxaca. All specimens known, including referred specimens, are from within this radius, and all are restricted to one biogeographical formation as defined by Grünwald et al. (2015), namely the Sierra Juárez Pine-Oak Woodland (#32 on p. 409 in Grünwald et al. 2015).

Due to its limited distribution and potential endemism near the type-locality, we propose this species be provisionally classified as Endangered B1ab(iii), based on the IUCN Red List Criteria that its occurrence is less than 100 km^2, it occurs in only one threat-defined location, and there is ongoing decline in the extent and quality of its habitat due to small-scale cattle ranching, illegal logging, and varied agricultural practices.

Acknowledgements

First and foremost, we thank our field crew for their consistent and impressive efforts in the field, even under adverse conditions. Specifically, Cynthia Ramírez-González, Héctor Franz-Chávez, Iván Ahumada-Carrillo, and André Grünwald-Nuño, all of whom helped collect type material. Iván furthermore provided excellent drawings of the hands and feet of the new species. Others who have helped us in the field during the length of this project include Carmen Mendoza-Portilla, Ámbar Lanomy Grünwald-Nuño, Ricardo Ramírez-Chaparro, Brandon La Forest, Jason M. Jones, Karen Itzel Morales-Flores, and many more who we are surely leaving out. We are equally indebted to María de los Ángeles Palma-Irizarry of the Secretaría de Medio Ambiente y Recursos Naturales (SEMARNAT) for providing collecting permits (SGPA/DGVS/002288/18). We thank Drs. Adrián Nieto Montes de Oca, Oscar Flores-Villela, and Gustavo Campillo from the Colección de Museo de Zoología, Facultad de Ciencias (MZFC) at UNAM; Drs. Gabriela Parra-Olea and Omar Hernández-Ordóñez at the Colección Nacional de Anfibios y Reptiles, Instituto de Biología (CNAR) at UNAM; and Dr. Ireri Suazo-Ortuño and Jonatan Torres-Pérez-Coeto from the Colección Herpetológica de la Universidad Michoacana (INIRENA) for receiving and cataloguing the type material. We want to thank the community of La Esperanza, Municipio de Santiago Comaltepec, who allowed us to collect frogs on their communal lands. Jesse Delia, Rogelio Simón-Salvador,

Angel Lara-Brenis, and Vicente Mata-Salva shared important data and photos of their observations on the new species in the field. Jason Strickland (University of Alabama) and Uri García-Vázquez (UNAM - FES Zaragoza) kindly shared sequences with us to include in our molecular analysis and Itzue Caviedes-Solis, Uri García-Vázquez and Zeltzin Corina Rodríguez-Ortíz shared trees from phylogenetic analyses they were performing which included *Charadrahyla*. Emily Braker from UCM, Enrique Santoyo-Brito from UIMNH, Ana Motta from KU, Erica Ely from CAS, and Gregory Pandelis from UTA all aided the project by kindly photographing specimens under their care. Ginny Weatherman and Ana Motta further assisted in photographing every single relevant specimen at KU. Finally, we thank Dr. Joseph Mendelson and Ricardo Palacios-Aguilar for reviewing an earlier version of this manuscript and their detailed commentary which improved the paper significantly; Joe Mendelson also generously shared his notes and measurements of *Charadrahyla* specimens at KU and UTA for our analysis and use.

We thank Biencom Real Estate, Biodiversa A.C. and Herp.mx A.C. for providing important funding for the fieldwork.

References

Adler K, Dennis DM (1972) New tree frogs of the genus *Hyla* from the cloudforests of western Guerrero, Mexico. Occasional Papers of The Museum of Natural History, University of Kansas 7: 1–19.

Altig RW, McDiarmid RW (1999) Development and Morphology. In: McDiarmid RW, Altig RW (Eds) Tadpoles: The Biology of Anuran Larvae. The University of Chicago Press, Chicago, Illinois, United States, 24–51.

Arreortúa M, Flores CA, Simón-Salvador PR, Santiago-Dionicio H, González-Bernal E (2021) Description of the tadpole and natural history notes of *Incilius spiculatus* (Mendelson, 1997), and Endangered toad endemic to the Sierra Madre de Oaxaca, Mexico. Amphibian & Reptile Conservation 15(2): 31–39.

Bautista del Moral A, Santos-Bibiano R, Cruz-Machuca TE, Reyes-García CM, Palacios-Aguilar R (2021) New locality records of the Mixteca cloudforest tree frog *Charadrahyla sakbah* from Guerrero, Mexico. The Herpetological Bulletin 155: 2021: 48–49. https://doi.org/10.33256/hb155.4849

Bossuyt F, Milinkovitch MC (2000) Convergent adaptive radiations in Madagascan and Asian ranid frogs reveal covariation between larval and adult traits. Proceedings of the National Academy of Sciences, USA 97: 6585–6590. https://doi.org/10.1073/pnas.97.12.6585

Caldwell JP (1974) Tropical tree frog communities: patterns of reproduction, size, and utilization of structural habitat. Unpublished Ph.D. Dissertation. University of Kansas, Lawrence, Kansas, USA. 197 pp.

Campbell JA, Lamar WW, Hillis DM (1989) A new species of diminutive *Eleutherodactylus* (Leptodactylidae) from Oaxaca, Mexico. Proceedings of the Biological Society of Washington 102: 491–499.

Campbell JA, Duellman WE (2000) New species of stream-breeding hylid frogs from the northern versant of the highlands of Oaxaca, Mexico. Scientific Papers. Natural History Museum, University of Kansas 16: 1–28.

Campbell JA, Blancas-Hernández JC, Smith EN (2009) A new species of stream-breeding treefrog of the genus *Charadrahyla* (Hylidae) from the Sierra Madre del Sur of Guerrero, Mexico. Copeia 2009(2): 287–295. https://doi.org/10.1643/CH-08-143

Canseco-Márquez L, Gutiérrez-Mayén G (2010) Anfibios y Reptiles del Valle de Tehuacán-Cuicatlán. CONABIO, Cuicatlán, A.C. Benemérita Universidad Autónoma de Puebla (BUAP), Puebla, Mexico, 302 pp.

Canseco-Márquez L, Ramírez-González CG, González-Bernal E (2017) Discovery of another new species of *Charadrahyla* (Anura, Hylidae) from the cloudforest of northern Oaxaca, México. Zootaxa 4329(1): 64–72. https://doi.org/10.11646/zootaxa.4329.1.2

Caviedes-Solis IW, Vázquez-Vega LF, Solano-Zavaleta I, Pérez-Ramos E, Rovito S, Devitt T, Heimes P, Flores-Villela OA, Campbell JA, Montes de Oca A (2014) Everything is not lost: recent records, rediscoveries and range extensions of Mexican hylid frogs. Mesoamerican Herpetology 2(3): 229–241.

Caviedes-Solis IW, Kim N, Leaché AD (2020) Species IUCN threat status level increases with elevation: a phylogenetic approach for Neotropical tree frog conservation. Biodiversity and Conservation 29(8): 2515–2537. https://doi.org/10.1007/s10531-020-01986-8

Delia JRJ, Whitney JL, Burkhardt T (2013) Rediscovery of 'lost' treefrogs from Oaxacan highlands of Mexico. Biodiversity Conservation 22(6): 1405–1414. https://doi.org/10.1007/s10531-013-0481-9

Duellman WE (1961) Description of a new species of tree frog from México. Studies of American Hylid Frogs VI. Herpetologica 17(1): 1–5. https://doi.org/10.2307/3626762

Duellman WE (1965) Frogs of the *Hyla taeniopus* Group. Copeia 1965(2): 159–168. https://doi.org/10.2307/1440718

Duellman WE (1968) Descriptions of new hylids frogs from Mexico and Central America. University of Kansas Publications, Museum of Natural History 17(13): 559–578. https://doi.org/10.5962/bhl.part.7138

Duellman WE (1970) The hylid frogs of Middle America. Monograph of the Museum of Natural History, University of Kansas 1: 1–753. [72 pls] https://doi.org/10.5962/bhl.title.2835

Duellman WE (2001) The hylid frogs of Middle America. Vol. 1 & 2. Society for the Study of Amphibians and Reptiles, Ithaca, NY, 603 pp. & 1159 pp. [92 pls]

Escobedo-Cadena RG, Hernández-Marmolejo LL, Grünwald CI, García-Vázquez UO (2024) Ampliación de la distribución de la rana *Charadrahyla taeniopus* (Anura: Hylidae) en el estado de San Luis Potosí, México. Revista Latinoamericana De Herpetología 7(2): 43–45 e821. https://doi.org/10.22201/fc.25942158e.2024.2.821

Faivovich J, Haddad CFB, Garcia PCA, Frost DR, Campbell JA, Wheeler WC (2005) Systematic review of the frog family Hylidae, with special reference to Hylinae: phylogenetic analysis and taxonomic revision. Bulletin of the American Museum of Natural History 294(1): 1–240. https://doi.org/10.1206/0003-0090(2005)294[0001:srotff]2.0.co;2]2.0.CO;2

Faivovich J, Pereyra MO, Luna MC, Hertz A, Blotto BL, Vásquez-Almazán CR, McCranie JR, Sánchez Da, Baêta D, Araujo-Vieira K, Köhler G, Kubicki B, Campbell JA, Frost DR, Wheeler WC, Haddad CFB (2018) On the monophyly and relationships of several genera of Hylini (Anura: Hylidae: Hylinae), with comments on recent taxonomic changes in Hylids. South American Journal of Herpetology 13(1): 1–32. https://doi.org/10.2994/SAJH-D-17-00115.1

Flores-Villela OA (1993) Herpetofauna Mexicana. Special Publications of the Carnegie Museum of Natural History 17: 1–73.

García-Padilla E, Villalobos-Juárez I, Bautista-Bautista L, Méndez-Hernández C, Torres-Barragán CA, Tomás-López C, López-Jiménez B, Martínez-Cruz JM, Sosa-Reyes M (2024) Nuevos

registros distribucionales de anfibios y reptiles en el estado de Oaxaca, México. Revista Latinoamericana de Herpetología 7(1): 137–142. [e878] https://doi.org/10.22201/fc.25942158e.2024.1.878

Gosner KL (1960) A simplified table for staging anuran embryos and larvae with notes on identification. Herpetologica 16(3): 183–190.

Grünwald CI, Jones JM, Franz-Chávez H, Ahumada-Carrillo IT (2015) A new species of *Ophryacus* (Serpentes: Viperidae: Crotalinae) from eastern Mexico, with comments on the taxonomy of related pitvipers. Mesoamerican Herpetology 2(4): 387–416.

Grünwald CI, Reyes-Velasco J, Franz-Chávez H, Morales-Flores KI, Ahumada-Carrillo IT, Jones JM, Boissinot S (2018) Six new species of *Eleutherodactylus* (Anura: systematic relationships and the validity of related species. Mesoamerican Herpetology 5(1): 6–83.

Günther CLGA (1901) Reptilia and Batrachia. In: Godman F, Salvin O (Ed.) Biologia Centrali-Americana, London, Zoology, 1–326.

Heimes P (2010) *Charadrahyla* – eine neue Laubfroschgattung aus Mexiko. Aquaristik-Fachmagazin 211/42(1): 80–88.

IUCN (2025) The IUCN Red List of Threatened Species. Version 2025-1. https://www.iucnredlist.org [Accessed on 28 July 2025]

Jiménez-Arcos VH, Calzada-Arciniega RA, Alfaro-Juantorena LA, Vázquez-Reyes LD, Blair C, Parra-Olea G (2019) A new species of *Charadrahyla* (Anura: Hylidae) from the cloudforest of western Oaxaca, Mexico. Zootaxa 4554: 371–385. https://doi.org/10.11646/zootaxa.4554.2.3

Katoh K, Rozewicki J, Yamada KD (2019) MAFFT online service: multiple sequence alignment, interactive sequence choice and visualization. Briefings in Bioinformatics 20(4): 1160–1166. https://doi.org/10.1093/bib/bbx108

Kumar S, Stecher G, Li M, Knyaz C, Tamura K (2018) MEGA X: Molecular Evolutionary Genetics Analysis across computing platforms. Molecular Biology and Evolution 35(6): 1547–1549. https://doi.org/10.1093/molbev/msy096

Lamoreux JF, McKnight MW, Cabrera-Hernández R (2015) Amphibian alliance for zero extinction sites in Chiapas and Oaxaca. Occasional Paper of the IUCN Species Survival Commission Number 53: xxiv + 1–320. https://doi.org/10.2305/iucn.ch.2015.ssc-op.53.en

León-Gómez A (2019) Efecto de la perturbación de bosque mesófilo de montaña sobre 4 especies de anuros nativos en la comunidad de la Esperanza, Oaxaca. Master's Thesis, Instituto Politécnico Nacional (CIIDIR, Oaxaca), Oaxaca, México, 79 pp.

López-Luna MA (2017) Estado actual de la diversidad y poblaciones de los anfibios y reptiles en Los Tuxtlas. In: Reynoso VH, Coates RI, Vázquez-Cruz ML(Eds) Avances y Perspectivas en la Investigación de los Bosques Tropicales y sus Alrededores: La Región de Los Tuxtlas. Instituto de Biología, Universidad Nacional Autónoma de México, Mexico City, Mexico, 347–369.

Lynch JD, Smith HM (1966) New or unusual amphibians and reptiles from Oaxaca, Mexico, II. Transactions of the Kansas Academy of Science 69: 58–75. https://doi.org/10.2307/3627636

Mata-Silva V, Rocha R, Mata-González S, Ramírez-Bautista A, Berriozabal-Islas C, García-Padilla E, Johnson JD, Wilson LD (2019) First municipality records and additional information on the herpetofauna of Oaxaca, Mexico. Herpetological Review 50(3): 533–536.

Mendelson JRIII, Campbell JA (1999) The taxonomic status of populations referred to *Hyla chaneque* in southern Mexico, with the description of a new treefrog from Oaxaca. Journal of Herpetology 33(1): 80–86. https://doi.org/10.2307/1565545

Myers CW, Duellman WE (1982) A new species of *Hyla* from Cerro Colorado, and other tree frog records and geographical notes from western Panama. American Museum Novitates 2752: 1–32.

Nguyen LT, Schmidt HA, Von Haeseler A, Minh BQ (2015) IQ-TREE: a fast and effective stochastic algorithm for estimating maximum-likelihood phylogenies. Molecular Biology and Evolution 32(1): 268–274. https://doi.org/10.1093/molbev/msu300

Palacios-Aguilar R, Ochoa-Ochoa LM, Colín-Martínez VH, Hernández-Rubio S (2022) New distributional records of the herpetofauna of southern and western Mexico. Herpetological Review 53(3): 449–450.

Pérez-Higareda G (1981) Geographic Distribution: Hyla Chaneque. Herpetological Review 12(2): 64.

Rambaut A (2014) FigTree v1. 4.2. http://tree.bio.ed.ac.uk/software/figtree/

Reyes-Velasco J (2024) A revision of recent taxonomic changes to the eyelash palm pitviper, *Bothriechis schlegelii* (Serpentes, Viperida). Herpetozoa 37: 305–318.

Ríos-Rodas L, Zenteno-Ruiz CE, Barragán-Vázquez MDR, Canseco-Márquez L, López-Luna MA (2019) New anuran records for Tabasco, Mexico. Checklist 15(6): 1161–1166. https://doi.org/10.15560/15.6.1161

Sabaj-Pérez HM (2016) Standard symbolic codes for institutional resource collections in herpetology and ichthyology: an Online Reference. Version 6.5 American Society of Ichthyologists and Herpetologists, Washington, D.C., United States. www.asih.org/ [accessed 13 March 2025]

Savage JM, Heyer RW (1997) Digital webbing formulae for anurans: a refinement. Herpetological Review 28(3): 131.

Simón-Salvador PR, Arreortúa M, Flores CA, Santiago-Dionicio H, González-Bernal E (2021) The role of indigenous and community conservation areas in herpetofauna conservation: a preliminary list for Santa Cruz Tepetotutla, Oaxaca, Mexico. ZooKeys 1029: 185–208. https://doi.org/10.3897/zookeys.1029.62205

Smith HM (1939) Mexican herpetological novelties. Proceedings of the Biological Society of Washington 52: 187–196.

Snyder DH (1972) *Hyla juanitae*, a new treefrog from southern México, and its relationship to *H. pinorum*. Journal of Herpetology 6(1): 5–15. https://doi.org/10.2307/1563087

Steindachner F (1864) Batrachologische Mittheilungen. Verhandlungen des Zoologisch- Botanischen Vereins in Wien 14: 239–288.

Stöck M, Dubey S, Klütsch CFC, Litvinchuk S, Scheidt U, Perrin N (2008) Mitochondrial and nuclear phylogeny of circum-Mediterranean tree frogs from the *Hyla arborea* Group. Molecular Phylogenetics and Evolution 49(3): 1019–1024. https://doi.org/10.1016/j.ympev.2008.08.029

Stuart SN, Hoffmann M, Chanson JS, Cox NA, Berridge RJ, Ramani P, Young BE (eds.) (2008) Threatened amphibians of the world. (2008): 241. Lynx Edicions, Barcelona, Spain; IUCN Gland, Switzerland; Conservation International, Arlington, Virginia, USA, 750 pp.

Taylor EH (1937) New species of hylid frogs from Mexico with comments on the rare *Hyla bistincta* Cope. Proceedings of the Biological Society of Washington 50: 43–54.

Taylor EH (1942) New tailless Amphibia from Mexico. University of Kansas Science Bulletin 28: 67–89. https://doi.org/10.5281/zenodo.16156946

Toal KR III, Mendelson JR III (1995) A new species of *Hyla* (Anura: Hylidae) from cloud forest in Oaxaca, Mexico, with comments on

the status of the *Hyla bistincta* Group. Occasional Papers of the Museum of Natural History, University of Kansas 174: 1–20.

Trifinopoulos J, Nguyen LT, von Haeseler A, Minh BQ (2016) W-IQTREE: a fast online phylogenetic tool for maximum likelihood analysis. Nucleic Acids Research 44(W1): W232–W235. https://doi.org/org/10.1093/nar/gkw256

Warwick AJ, Barrow LN, Smith ML, Means DB, Lemmon AR, Lemmon EM (2021) Signatures of north-eastern expansion and multiple refugia; genomic phylogeography of the Pine Barrens tree frog, *Hyla andersonii* (Anura: Hylidae). Biological Journal of the Linnean Society 133(1): 120–134. https://doi.org/10.1093/biolinnean/blab023

Wiens JW, Kuczynski CA, Hua X, Moen, DS (2010) An expanded phylogeny of treefrogs (Hylidae) based on nuclear and mitochondrial sequence data. Molecular Phylogenetics and Evolution 55(3): 871–882. https://doi.org/10.1016/j.ympev.2010.03.013

Wüster W (2025) Shedding the mitochondrial blinkers: A long-overdue challenge for species delimitation in herpetology. Vertebrate Zoology 75: 259–275. https://doi.org/10.3897/vz.75.e161536

Yan P, Pan T, Wu G, Kang X, Ali I, Zhou W, Li J, Wu X, Baowei Z (2020) Species delimitation and evolutionary history of tree frogs in the *Hyla chinensis* Group (Hylidae, Amphibian). Frontiers in Ecology and Evolution 8: 234. https://doi.org/10.3389/fevo.2020.00234

Zhang P, Liang D, Mao RL, Hillis D, Wake D, Cannatella D (2013) Efficient sequencing of anuran mtDNAs and a mitogenomic exploration of the phylogeny and evolution of frogs. Molecular Biology and Evolution 30: 1899–1915. https://doi.org/10.1093/molbev/mst091

Appendix 1

Specimens examined:

***Charadrahyla altipotens*: MEXICO: OAXACA:** 37 km N of San Gabriel Mixtepec, Municipio de San Juan Lachao (CIG 1018–19); km. 179 on Hwy 175 between Oaxaca and Candelaria Loxicha, Municipio de San Agustin Loxicha (CIG 1234).

***Charadrahyla chaneque*: MEXICO: CHIAPAS:** (CIG 1313, CIG 2420); Selva Negra, Municipio de Rayón Mescalapa; **OAXACA:** Chalchijapa, Santa María Chimalapa (MZFC 18911–12); 6.2 km S of Rayón 6.4 km S of Rayón Mescalapa, Municipio de Rayón Mescalapa, Municipio de Rayón Mescalapa (KU 58439 [holotype], photo only); **VERACRUZ:** Cumbre de Santa Marta, Municipio de Soteapan (CNAR 18933).

***Charadrahyla "duellmani" aff. chaneque*: MEXICO: OAXACA:** Sierra Madre north of Zanatepec, 5,000 ft., (UIMNH 56812[holotype]; KU 106296, photo only).

***Charadrahyla esperancensis*: MEXICO: OAXACA:** Arroyo Los Tres Manantiales, La Esperanza, Municipio de Santiago Comaltepec (MZFC 28699); Hwy. 175, near La Esperanza, Municipio de Santiago Comaltepec (MZFC 28693).

***Charadrahyla juanitae*: MEXICO: GUERRERO:** 1.6 km SE of San Andrés de la Cruz, Municipio de Atoyac de Álvarez (CIG 1477–78); small stream immediately south of Santa Catarina Juquila, on rd. to Puerto Escondido, Municipio de Santa Catarina Juquila (CIG 0636–37).

***Charadrahyla nephila*: MEXICO: OAXACA:** 5.8 km W of Totontepec, headwaters of the Río Lana, Municipio Totontepec Villa de Morelos (UTA 5768[holotype] photo only), (UTA 5770=CNAR 28053, UTA 5784, UTA 5785=CNAR 28052, photo only); 5.0 km W of Totontepec, Municipio de Totontepec Villa de Morelos (UTA 6841–42, 6934–35, photo only); 4.2 km S of Vista Hermosa, Municipio de Santiago Comaltepec (KU 86961–65, 100900–01, 152365, photo only); 5.0 km S of Vista Hermosa, Municipio de Santiago Comaltepec (KU 58445, photo only); Arroyo Buena Vista, 6.5 km S of Vista Hermosa (KU 71123–26, photo only); stream crossing Hwy 175, 7.2 km S Vista Hermosa, Municipio de Santiago Comaltepec (MZFC 16796, 16822); 7.5–7.8 km S of Vista Hermosa, Municipio de Santiago Comaltepec (KU 86956, 136661, photo only); 9.1–9.4 km S of Vista Hermosa, Municipio de Santiago Comaltepec (KU 136662–65, photo only); 11.1–11.9 km S of Vista Hermosa, Municipio de Santiago Comaltepec (KU 86968, 136666–85, photo only); 12.3 km S of Vista Hermosa, Municipio de Santiago Comaltepec (136686, 136688, photo only); 12.8 km S of La Esperanza, Municipio de Santiago Comaltepec (MZFC 37340–43); 13 km S of Vista Hermosa, Municipio de Santiago Comaltepec (KU 100899), 15.0–16.0 km S of Vista Hermosa, Municipio de Santiago Comaltepec (KU 86971, 136689–91, photo only); 16.6 km S of Vista Hermosa, Municipio de Santiago Comaltepec (INIRENA 2951, KU 136692–93, photo only); Carretera Totontepec-Ayutla, Municipio de Totontepec Villa de Morelos (MZFC 37338–39); 6.4 km S of Vista Hermosa Municipio de Santiago Comaltepec (UCM 41323–24, photo only); 12.8 km S of Vista Hermosa, Municipio de Santiago Comaltepec (UCM 41272–75, photo only); 15.4 km N of Cerro Pelón, Municipio de Santiago Comaltepec (UTA 4770, photo only); 16.1 km N of Cerro Peón, Municipio de Santiago Comaltepec (UTA 4769, photo only); 33.9 km S of Valle Nacional, Municipio de Santiago Comaltepec (UTA 3074–75, photo only); 12.2 km S of Vista Hermosa, Municipio de Santiago Comaltepec (UTA 54901–02, photo only); 4 km S of Vista Hermosa (UTA 6836, photo only); 16 km S of Vista Hermosa, Municipio de Santiago Comaltepec (UIMNH 57195–96, photo only); La Luz Yagila, Municipio de Ixtlán de Juárez (CIG 02609); 40.1 km E of Teotitlán, Sierra Mazateca, Municipio de Santa María Teopoxco (UTA 13194–95, photo only); Plan de Guadalupe, Sierra Mazateca, Municipio de Santa María Teopoxco (CIG 2505).

***Charadrahyla pinorum*: MEXICO: GUERRERO:** 1.6 km SE of San Andrés de la Cruz, Municipio de Atoyac de Álvarez (CIG 1480).

***Charadrahyla sakbah*: MEXICO: OAXACA:** Río Chite ku'e (Río de las Mil Cascadas), San Isidro Paz y Progreso, Municipio de Santa Maria Yucuhiti (CNAR 30989–94); stream on Cerro Monteverde, approximately 12.6 km (airline) NE of Zoquiápam Boca de los Ríos, Municipio de San Juan Bautista Atatlahuca (MZFC 21980); stream on Cerro Monteverde, approximately 11.7 km (airline) NE of Zoquiápam Boca de los Ríos, Municipio de San Juan Bautista Atatlahuca (MZFC 21978–79; 21981–82).

***Charadrahyla taeniopus*. MEXICO: HIDALGO**: Apantlazol, 4 km E of Tlanchinol, Municipio de Tlanchinol (MZFC 6484, 6485, 21036); El Temapá, Municipio de Tenango de Doria (MZFC 6486), 1 km NE Zetoy (MZFC 7713), El Texme, Municipio de Tenango de Doria (MZFC 7714); dirt road to Eloxochitlán, Municipio de Eloxochitlán (MZFC 21035); Huehuetla, Municipio de Huehuetla (MZFC 7715); Bernardo, 1.5 km (MZFC 6453), Tlanchinol, Municipio de Tlanchinol (MZFC 15235), Just S of Calnali, Municipio de Canali (MZFC 21034); **PUEBLA:** Río Xoloatl, 5 km NE of Teziutlán, Municipio de Teziutlán (MZFC 5149-56); Sompatisoya, Municipio de Hueytamalco (MZFC 8297), Villas Cuetzalan, Municipio de Cuetzalan de Progreso (MZFC 16301-03); Rancho Las Truchas, Municipio de Huauchinango (CIG 1647) **SAN LUIS POTOSÍ:** Cueva de los Manantiales 2, Valle de Trinidad, Municipio de Xilitla (CIG 1105 –06); La Silleta, Municipio de Xilitla (MZFZ 4482); **VERACRUZ:** Los Capulines, Municipio de Yecuautla (CIG 1383–84, 1400); Puente Ruiz Cortines, Municipio de Huatusco (CIG 2235–36).

***Charadrahyla trux*: MEXICO: GUERRERO:** 11.4 km (by road) southwest of Puerto del Gallo, Municipio de Atoyac de Álvarez (KU 137551); 10.4 km SW of Puerto del Gallo, Municipio de Atoyac de Álvarez (KU 137550).

***Charadrahyla viruela*: MEXICO: OAXACA:** Highway 175 between Vista Hermosa and La Esperanza, 1.9 km ENE of La Esperanza, Municipio de Santiago Comaltepec (INIRENA 2950; MZFC 37344); Arroyo Tres Manantiales, La Esperanza, Municipio de Santiago Comaltepec (MZFC 37345); Arroyo Tepetotutla, arboles pivotantes, Municipio de San Felipe Usila, (CNAR 24781, 24808); Puente San Isidro, Municipio de San Pedro Yolox (CNAR 24782); 4.2 km S Vista Hermosa on Hwy 175, ca 1.2 km S La Esperanza, Municipio de Santiago Comaltepec (MZFC 15974–75, INIRENA 2948–49; CIG 2610; KU 86966, 136656–59 photo only); 8 km S of Yetla, Municipio de San Juan Bautista Valle Nacional (KU 86967, photo only); 9 km S of Yetla, Municipio de San Juan Bautista Valle Nacional (KU 64355, 136651, 136655, photo only).

Appendix 2

Table A1. Collection and voucher data for genetic samples of *Charadrahyla* used in this study, either to create the phylogenetic tree or to calculate genetic distances, or both. Acronyms for herpetological collections follow Sabaj-Pérez (2016). JAC, CIG, JCBH and JJW, are field identifiers for un-catalogued specimens being deposited in the INIRENA, MZFC, UTA. New sequences are indicated in bold.

No.	Voucher number	Taxa	Locality	12S	16S	ND1	RAG1	Rhodopsin
1	CIG_1234	*Charadrahyla altipotens*	Mexico: Oaxaca: San Gabriel Mixtepec	**PZ143181**	**PZ138301**	–	–	–
2	ICS_680	*Charadrahyla altipotens*	Mexico: Oaxaca: San Gabriel Mixtepec	–	Caviedes-Solis et al. 2020	–	Caviedes-Solis et al. 2020	Caviedes-Solis et al. 2020
3	SMF_100340	*Charadrahyla altipotens*	Mexico: Oaxaca: Santa María Jalatengo	–	KX423495	–	–	–
4	INIRENA_2954	*Charadrahyla chaneque*	Mexico: Chiapas: Rayón Mescalapa	**PZ143186**	–	–	–	–
5	MZFC_28699	*Charadrahyla esperancensis*	Mexico: Oaxaca: Santiago Comaltepec	–	Caviedes-Solis et al. 2020	–	Caviedes-Solis et al. 2020	Caviedes-Solis et al. 2020
6	CIG_1477	*Charadrahyla juanitae*	México: Guerrero: San Vicente de Benítez	**PZ143182**		–	–	–
7	IBH_27011	*Charadrahyla juanitae*	Mexico: Oaxaca: San Gabriel Mixtepec	–	KX423491	–	–	–
8	ICS_647	*Charadrahyla juanitae*	Mexico: Oaxaca	–	Caviedes-Solis et al. 2020	–	Caviedes-Solis et al. 2020	Caviedes-Solis et al. 2020
9	SMF_96516	*Charadrahyla juanitae*	Mexico: Oaxaca: Buenavista Loxicha	–	KX423493	–	–	–
10	JAC_21531	*Charadrahyla nephila*	México: Oaxaca: Totontepec	**PZ143187**	AY843649	–	AY844438	AY844635
11	CIG_1480	*Charadrahyla pinorum*	Mexico: Guerrero: San Vicente de Benítez	**PZ143183**	–	–	–	–
12	VHJA_0242	*Charadrahyla sakbah*	Mexico: Oaxaca: San Isidro Paz y Progreso	–	Caviedes-Solis et al. 2020	–	Caviedes-Solis et al. 2020	Caviedes-Solis et al. 2020

No.	Voucher number	Taxa	Locality	12S	16S	ND1	RAG1	Rhodopsin
13	MZFC_15720	*Charadrahyla taeniopus*	Mexico: Puebla: Huauchinango	AY819455	AY819556	–	–	–
14	JAC_24443	*Charadrahyla taeniopus*	Mexico: Puebla: Cuetzalan	AY843679	AY843679	MH004120	AY844463	AY844671
15	JCBH_1000	*Charadrahyla trux*	Mexico: Guerrero: Atoyac	–	Caviedes-Solis et al. 2020	–	Caviedes-Solis et al. 2020	Caviedes-Solis et al. 2020
16	INIRENA_2950	*Charadrahyla viruela* sp. nov.	Mexico: Oaxaca: Santiago Comaltepec	**PZ143185**	**PZ138302**	–	–	–
17	INIRENA_2949	*Charadrahyla viruela* sp. nov.	Mexico: Oaxaca: Santiago Comaltepec	**PZ143184**	–	–	–	–
18	JAC_21583	*Megastomatohyla mixe*	Mexico: Oaxaca: Santiago Zacatepec	AY843646	AY843646	–	AY844434	–
19	ICS_526	*Megastomatohyla mixomaculata*	Mexico: Oaxaca: Sierra Mazateca	–	Caviedes-Solis et al. 2020	–	–	–
20	MZFC_28696	*Megastomatohyla mixomaculata*	Mexico: Puebla: San Sebastian Tlacotepec	–	**PZ138303**	–	–	–
21	IBH_27027	*Megastomatohyla pellita*	Mexico: San Gabriel Mixtepec	MH004093	KX423513	–	–	–
22	SMF_96479	*Megastomatohyla pellita*	Mexico: San Gabriel Mixtepec	–	**PZ138304**	MH004145	–	–

Supplementary material 1

12S Phylogenetic Tree

Authors: Jacobo Reyes-Velasco

Data type: pdf

Explanation note: Maximum Likelihood phylogenetic inference of members of the genus *Charadrahyla* and *Megastomatohyla*, based on the mitochondrial gene 12S.

Copyright notice: This dataset is made available under the Open Database License (http://opendatacommons.org/licenses/odbl/1.0/). The Open Database License (ODbL) is a license agreement intended to allow users to freely share, modify, and use this Dataset while maintaining this same freedom for others, provided that the original source and author(s) are credited.

Link: https://doi.org/10.3897/herpetozoa.39.e184954.suppl1

Supplementary material 2

16S Phylogenetic Tree

Authors: Jacobo Reyes-Velasco

Data type: pdf

Explanation note: Maximum Likelihood phylogenetic inference of members of the genus *Charadrahyla* and *Megastomatohyla*, based on the mitochondrial gene 16S.

Copyright notice: This dataset is made available under the Open Database License (http://opendatacommons.org/licenses/odbl/1.0/). The Open Database License (ODbL) is a license agreement intended to allow users to freely share, modify, and use this Dataset while maintaining this same freedom for others, provided that the original source and author(s) are credited.

Link: https://doi.org/10.3897/herpetozoa.39.e184954.suppl2

Supplementary material 3

12S Genetic Distance Table

Authors: Jacobo Reyes-Velasco

Data type: xlsx

Explanation note: Genetic distance table of the genetic distances of 12S mitochondrial gene for various samples of *Charadrahyla* and related Hylidae.

Copyright notice: This dataset is made available under the Open Database License (http://opendatacommons.org/licenses/odbl/1.0/). The Open Database License (ODbL) is a license agreement intended to allow users to freely share, modify, and use this Dataset while maintaining this same freedom for others, provided that the original source and author(s) are credited.

Link: https://doi.org/10.3897/herpetozoa.39.e184954.suppl3

Supplementary material 4

16S Genetic Distance Table

Authors: Jacobo Reyes-Velasco

Data type: xlsx

Explanation note: Genetic distance table of the genetic distances of 16S mitochondrial gene for various samples of *Charadrahyla* and related Hylidae.

Copyright notice: This dataset is made available under the Open Database License (http://opendatacommons.org/licenses/odbl/1.0/). The Open Database License (ODbL) is a license agreement intended to allow users to freely share, modify, and use this Dataset while maintaining this same freedom for others, provided that the original source and author(s) are credited.

Link: https://doi.org/10.3897/herpetozoa.39.e184954.suppl4